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The Elisha Mitchell Scientific Society

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Journal of the Elisha Mitchell Scientific Society

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In addition to original papers on scientific subjects, this Journal publishes the Proceedings of the Elisha Mitchell Scientific Society, the Proceedings of the North Carolina Academy of Science, and abstracts of papers read before the North Carolina Bacteriological Society and the North Carolina Psychological Association, including some abstracts of papers published elsewhere.

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PROCEEDINGS AND ABSTRACTS FROM THE FIFTY-EIGHTH ANNUAL MEETING OF THE NORTH CAROLINA ACADEMY OF SCIENCE, MAY 5–6, 1961, AT RALEIGH, N. C.

The fifty-eighth annual meeting of the North Carolina Academy of Science was held at North Carolina State College, Raleigh, on May 5 and 6, 1961.

The Academy met in its first session at 10:15 a.m. on May 5 with President Jensen presiding. A general session was held on this occasion, the papers coming from several academy members.

The business meeting of the Academy was held at 4:30 p.m. on May 5, at which time the following business was transacted.

The minutes of the 1960 meeting were approved as published in the *Journal of the Elisha Mitchell Scientific Society* **76**: 169–194.

Report of the Executive Committee

The Executive Committee met in its spring meeting at North Carolina State College on May 4. At this meeting the following matters were considered:

1. All persons who had applied for membership to this date were declared elected. Names of all new members appear later in the Proceedings.
2. Voted life membership in the Academy to O. C. Bradbury of Wake Forest College, Eva G. Campbell of Guilford College, B. B. Brandt of North Carolina State College, and Helen Barton of the Woman's College of the University of North Carolina.
3. Approved the following steering committee for the preparation of proposals for National Science Foundation grants: H. E. Speece, chairman; H. A. Shannon; and J. A. Yarbrough.
4. Selected Charles M. Allen of Wake Forest College as our representative on the AAAS Council and the Academy Conference for the 1961 meeting at Denver. Voted an allowance of $100 toward his expenses.
5. Voted to continue the payment of $1.00 per member together with an additional sum of $300 to the *Journal of the Elisha Mitchell Society* for 1960–61.
6. Voted to appropriate $50 for an additional Derieux Award in the Collegiate Academy for 1961. This was made necessary because of the large number of papers to be presented in the Collegiate Academy.
7. Voted to elect an editor as an additional officer in the North Carolina Academy of Science. A slight amendment to the Constitution which names the editor as an officer for a term of three years was approved.
8. In response to a letter from Dr. Dael Wolfle of the AAAS, voted to ask that the AAAS continue its allotment of research money to the respective Academies on the same basis as in past years.
9. Voted to accept the invitation of Wake Forest College for the 1962 annual meeting of the Academy. The dates of May 4 and 5, 1962, were set for the meeting.
10. The following were elected to membership in the Academy or reinstated to active membership:

Adamson, Claude L., Warren Wilson College, Swannanoa

Alliston, Dr. Charles Walter, North Carolina State College, Raleigh

Apple, Dr. Jay Lawrence, North Carolina State College, Raleigh

Binkley, Lowell E., Lenoir Rhyne College, Hickory

Brehme, Dr. Robert W., Wake Forest College, Winston-Salem

Butler, James Robert, University of North Carolina, Chapel Hill

Centini, Barry A., North Carolina State College, Raleigh

Cooper, Dr. Arthur Wells, North Carolina State College, Raleigh

Crutchfield, Philip J., University of North Carolina, Chapel Hill

Davis, John B., Jr., East Carolina College, Greenville

Davis, Dr. Robert Lloyd, University of North Carolina, Chapel Hill

Dawson, John W., Army Research Office, Durham
Eller, Dr. Frank W., East Carolina College, Greenville
Franklin, Mrs. Mary Ann W., Elizabeth City State Teachers College, Elizabeth City
Furbish, William J., Duke University, Durham
Goulson, Dr. Hilton T., University of North Carolina, Chapel Hill
Gregory, Dr. M. Pfluge, North Carolina State College, Raleigh
Gutierrez, Joseph Andres, Vulcan Materials Co., Winston-Salem
Gutknecht, John William, University of North Carolina, Chapel Hill
Haasis, Dr. Frank A., North Carolina State College, Raleigh
Haisley, Dr. Waldo Emerson, University of North Carolina, Chapel Hill
Hanahan, John, Jr., Belmont Abbey College, Belmont
Harris, Thomas M., University of North Carolina, Chapel Hill
Haynes, Dr. Frank L., Jr., North Carolina State College, Raleigh
Henderson, Dr. Warren R., North Carolina State College, Raleigh
Hester, Dr. F. Eugene, North Carolina State College, Raleigh
Hirt, Julian, Western Carolina College, Cullowhee
Hubbell, Davis Heuston, North Carolina State College, Raleigh
Humm, Dr. Douglas G., University of North Carolina, Chapel Hill
Leonard, Bobby Eugene, The Research Triangle Institute, Durham
McClure, Dr. Claude, University of North Carolina, Chapel Hill
McCombs, Dr. C. L., North Carolina State College, Raleigh
McDonald, Dr. James Clifton, Wake Forest College, Winston-Salem
Miller, J. Alfred, Bell Telephone Laboratories, Winston-Salem
Mueller, Miss Anna M., University of North Carolina, Chapel Hill
Murphy, Ted Daniel, Duke University, Durham
Nelson, Elijah L., Campbell College, Buies Creek
Nicely, Kenneth A., North Carolina State College, Raleigh
Norman, Dr. Vello, Liggett & Myers Tobacco Co., Durham
Olive, Dr. Aulsey Thomas, Wake Forest College, Winston-Salem
Parks, Clifford Robert, North Carolina State College, Raleigh
Paris, Dr. Oscar Hall, University of North Carolina, Chapel Hill
Peek, Harry M., North Carolina Department of Water Resources, Raleigh
Pinschmidt, Mrs. Mary Warren, Duke University, Durham
Powers, Dr. Maurice C., Catawba College, Salisbury
Prather, Mrs. Audrey M., Elizabeth City State Teachers College, Elizabeth City
Prichard, Dr. Robert W., Bowman Gray School of Medicine, Winston-Salem
Proffit, Mrs. G. T., Campbell College, Buies Creek
Rand, Miss Patricia J., Duke University, Durham
Ridley, Dr. Walter N., Elizabeth City State Teachers College, Elizabeth City
Sisler, Dr. Edward Carroll, North Carolina State College, Raleigh
Slade, Dr. Brenda Faulkner, Otago University, Dunedin, New Zealand
Stallworth, L. N., Elizabeth City State Teachers College, Elizabeth City
Steward, Robert F., Western Carolina College, Cullowhee
Taylor, Joseph R., Mars Hill College, Mars Hill
Taylor, Y. A., Department of Public Instruction, Raleigh
Thomas, Mrs. Dorothy Elliott, Elizabeth City State Teachers College, Elizabeth City
Ulberg, Dr. L. C., North Carolina State College, Raleigh
Vergeer, Dr. T., Appalachian State Teachers College, Boone
Watson, Andrew C., Warren Wilson College, Swannanoa
Watson, Dr. R. G., Bowman Gray School of Medicine, Winston-Salem
Wheeler, Dr. Walter H., University of North Carolina, Chapel Hill
Williams, Paul Lyle, Jr., Warren Wilson College, Swannanoa
Yang, Miss Shung-jun, North Carolina State College, Raleigh

Patron members as of July 1, 1961

W. Y. Preyer, Greensboro
C. M. Goethe, Sacramento, California
Richard E. Thigpen, Charlotte
Harold D. Cooley, House of Representatives, Washington, D. C.
Justice Emery B. Denny, Raleigh
C. P. Street, Charlotte
Irving E. Carlyle, Winston-Salem
Edward R. Zane, Greensboro

Sustaining members as of July 1, 1961

E. I. du Pont de Nemours & Company, Kinston
The Dickson Foundation, Inc., Mount Holly
Liggett & Myers Tobacco Company, Durham
Celanese Corporation of America, Charlotte
North Carolina Pharmaceutical Foundation, Inc., Chapel Hill

The above reports and actions of the Executive Committee were approved by the Academy in its business session of May 5, 1961.

The Poteat Award

The award for this year was given to the best paper presented before the Physics Section. The award was won by Dr. Wesley O. Doggett of North Carolina State College for his paper entitled A New Method of Determining the True Counting Rate from a Nuclear Detector. A certificate will be presented at the Academy banquet next year, 1962, at the meeting in Winston-Salem.

Report of the Research Committee

The following policies and procedures with respect to the award of research grants were adopted by the committee:

In the fall an announcement will be sent out to the various science departments in colleges and universities of the state of the availability of modest funds from the Academy for the support of research projects of *undergraduate* students. December 15 will be the deadline for receipt of applications, which will be considered sometime after January 1, when the amount of money available to the committee should be known.

A second announcement will be sent out sometime during the spring, depending on remaining funds, and at that time applications will be received from both undergraduate and graduate students. This procedure with respect to deadlines was established in order to encourage greater participation by undergraduate students. In both announcements a statement will be made asking that requests remain small and realistic in keeping with genuine need. No maximum will be stated, but in general the committee believes that grants above $200 should not be made except under very unusual circumstances. The feeling was that a stated maximum would encourage individuals to make requests up to this maximum when they might otherwise ask for less.

The cost of travel by private automobile will be calculated at five cents per mile.

Earlier in the year the committee decided that a final report would be requested from each recipient of a research grant in which a statement is given of the actual expenditure of grant funds. Recipients are also encouraged to report the results of their research at an annual Academy meeting.

For the coming year Dr. John Parker has agreed to serve as chairman of the committee. Both Dr. Berkut and Dr. Cutter have plans which will take them out of the state during a part or all of this period. They have agreed to continue to serve on the committee during this period, by correspondence if necessary. Dr. Jenner has agreed to assist at least to a limited degree in the work of the committee for the coming year, even though his term has expired. Dr. John Vernburg is the new member of the committee.

The following grants were made during 1960–61: Barry Centini, N. C. State College, $50; Donald Swift, U. N. C., $200; Kenneth Nicely, N. C. State College, $300; James Parnell and John Paul, N. C. State College, $200; Anna M. Mueller, U. N. C., $100; James A. Miller, N. C. State College, $90; and Mrs. Anne McCrary, U. N. C., $160.

C. E. JENNER, *Chairman*

Report of the Conservation-Legislative Committee

1. A "Memorandum of Understanding" has been consummated with Mr. Paul K. Camp in which the best portion of the white pine area on the Rocky River in Chatham County is being preserved and protected. This was done through the efforts of Dr. C. F. Korstian.

2. Negotiations are under way with Mr. J. T. Marks for a similar preservation of the white pine and mixed mesophytic area on the Deep River in Lee County. Mr. Marks will not give or sell the land at present.

3. A reconnaissance and preliminary negotiations have been made for the preservation of a hardwood forest over marl on Island Creek in Jones County. Dr. Frank Woods of Nature Conservancy is pushing this project.

4. A reconnaissance has been made for a suitable savannah site for preservation. Part of the Burgaw savannah is still intact, and an effort will be made to preserve this site unless a a more satisfactory area is found.

5. Through the alertness and work of Dr. F. S. Barkalow, action was taken for the defeat of the leasing of a microwave tower site in Mt. Jefferson State Park in Ashe County. A resolution written by Dr. Barkalow accompanied

by a letter from the committee was sent to each member of the Council of State. Mr. Hargrove Bowles, Director of the Department of Conservation and Development, has recently notified the committee of the withdrawal of the lease application by the Central Telephone Company.

6. An effort has been made to discuss the status of the "Venus fly-trap law" and other conservation matters with Mr. Hargrove Bowles. No mutually satisfactory time has been arranged for the discussion.

C. F. KORSTIAN; T. L. QUAY; L. M. OUTTEN; A. E. RADFORD, *Chairman*

A Resolution on State Parks

WHEREAS our state parks represent areas of special importance to all the people of North Carolina;

AND WHEREAS these areas were originally set aside to perpetuate and protect areas of unique beauty and scientific interest for both present and future generations of North Carolinans;

AND WHEREAS areas thus set aside represent some of the last vestiges of our once bountiful heritage of unexploited natural resources;

AND WHEREAS the maintenance of the integrity of all our state parks, free from the dangers of encroachment and exploitation, is essential if they are to fulfill their intended purpose;

AND WHEREAS equally advantageous alternate sites are available outside our state parks for private and/or public utility development and for the construction of flood control structures;

NOW, THEREFORE, BE IT RESOLVED that the North Carolina Academy of Science go on record as opposed to the impairment, destruction, or exploitation of any state park by public or private interests except in the case of dire national emergency; and furthermore be it recorded that the North Carolina Academy of Science is opposed to leasing of publicly owned state park lands to private organizations for private use and personal gain.

FURTHERMORE, BE IT RESOLVED that the North Carolina Academy of Science go on record as being opposed to the construction of flood-control structures in state parks whether for the protection of public or private lands.

BE IT FURTHER RESOLVED that the North Carolina Academy of Science express its appreciation to the officials of the State of North Carolina for their past, present, and future interest in furthering the development of our state parks system and for protecting the integrity of the parks from impairment, destruction, exploitation, and incompatible park uses.

BE IT FURTHER RESOLVED that the President of the North Carolina Academy of Science be directed to forward a copy of this resolution to the Governor of North Carolina, to each member of the Council of State, to the Director of the Department of Administration, to the Director and each member of the Board of Conservation and Development.

A Resolution on Mount Jefferson State Park

WHEREAS our state parks represent areas of special importance to all the people of North Carolina;

AND WHEREAS these areas were originally set aside to perpetuate and protect areas of unique beauty and scientific interest for both present and future generations of North Carolinians;

AND WHEREAS areas thus set aside represent some of the last vestiges of our once bountiful heritage of unexploited natural resources;

AND WHEREAS the maintenance of the integrity of all our state parks, free from the dangers of encroachment and exploitation, is essential if they are to fulfill their intended purpose;

AND WHEREAS the construction of a microwave tower in Mount Jefferson State Park would have been detrimental to this park, and whereas the leasing and construction of this tower would have set a dangerous precedent for all state parks;

AND WHEREAS the Central Telephone Company of Hickory after due deliberation and consideration has voluntarily withdrawn its request for a lease and permission to construct a microwave tower in Mount Jefferson State Park;

NOW, THEREFORE, BE IT RESOLVED that the North Carolina Academy of Science commend the Central Telephone Company of Hickory for its action in withdrawing its request.

BE IT FURTHER RESOLVED that the President of the North Carolina Academy of Science be directed to forward a copy of this resolution to the President of the Central Telephone Company of Hickory and to the Governor of North Carolina, to each member of the Council of State, to the Director of the Department of Administration, to the Director and each member of the Board of Conservation and Development.

Report of the Collegiate Academy

The two most successful events in the Collegiate Academy calendar this year were the annual meeting and the spring field trip.

At the annual meeting held Friday afternoon, May 5, at North Carolina State College, thirteen papers were presented, again necessitating two sections. The Derieux award for the Biological Science Section was awarded to W. Keith Baucom and Kenneth M. Lewis of North Carolina State College and for the Physical Science Section to Arlan Mantz of Catawba College. Talmadge T. Brown, Jr., of North Carolina State College and Mrs. Anne McCrary were given honorable mention in the Biological Seience Section, and James O. Rice and Walter J. Harrison of Agriculture & Technical College received honorable mention in the Physical Science Section. At the business meeting that followed, the availability of money from the senior academy to help finance research was stressed, and students were urged to apply for grants. Election of officers followed and in the evening a social hour was held from 6:30 to 8:00. Dr. John A. Santolucito was in charge of the annual meeting this year.

On April 22, 1961, over 100 students from eight colleges arrived at Beaufort for a spring collecting trip. Most of them came Friday night and attended the evening session at the Duke University Marine Laboratory, where they were divided into groups and given general information by the president, Mrs. Ruby Blackmore, and by Dr. Vernberg. Saturday morning the groups assembled early for the various field trips, after which the collections were spread out in the laboratories and identified.

In the lectureship program, which is managed by Dr. F. John Vernberg, sixteen speakers spoke to groups at eleven different colleges this year. We feel that this is an opportunity for science clubs to secure good speakers which should be more extensively used.

Two issues of CANCAS were published this year containing abstracts of most of the papers given at last year's annual meeting. We are indebted to the Sigma Xi chapters of the state for contributions which finance this publication.

Our membership this year consisted of thirteen clubs and twenty individual members.

CHARLOTTE DAWLEY

North Carolina Science Fair Program

In May of 1960, Dr. H. D. Crockford retired as the director of the North Carolina Science Fair Program, and Mr. A. F. Jenzano was appointed his successor. The program was activated with a meeting of the Planning and Policy Committee on October 18, 1960. Related practices, procedures, and policies were discussed, clarified, compiled, and distributed to all district directors.

In January of 1961, publicity packets consisting of descriptive science-fair materials and an appeal for consistent newspaper support were distributed to the editors of all North Carolina daily and weekly newspapers. Publicity about these activities stimulated early inquiries about the fairs. Inquiries were answered by the recipient directors or forwarded by categories to other professional men for accurate and helpful reply. All color slides (of previous exhibits) were recalled by mail and reapportioned together with a new set of the 1960 exhibit entries slides to each district director for "loan distribution."

A promising innovation this year was a weekly 30-minute science fair WUNC television show conducted by the director each Thursday evening from February 9 through April 13. Dr. Homer C. Folks made advance arrangements for two or three students and a science teacher from nearby schools to participate (with projects material) each week. Both the participants and verbal and written statements from viewers attested to the fruitfulness of the program, and every effort should be made to continue this practice in future years. However, if the series can be *scheduled* as early as January 15 to begin in mid-February, the advance time will permit better planning for more effective programming. Complete records of each program presented are in the science fair program files.

The state fair, held at the Men's Graduate Center of Duke University on April 7–8, was one of the finest and best organized since the inception of the North Carolina Science Fair program. Our 1960–61 state winners were:

1. Margaret Green, Enka High School, with her exhibit Taxonomy of *Cladonia cristatella.*
2. John C. Theys, Jr., Cary High School, with his exhibit Semiautomatic Binary Digital Computer.

At the national fair in Kansas City Miss Green won third-place honors in the Biological Division in competition with 385 winners from other states and countries entered.

Exhibits at all levels this year appeared to be of higher academic and presentational quality, expecially in the Junior Divison.

A. F. JENZANO, *Director*

Seventh Annual North Carolina Talent Search

In 1961 663 examinations were requested for Westinghouse Science Talent Search in North Carolina; 83 entries, which included a completed examination, project write up, and recommendations of a science teacher, were returned. Three applicants received national honorable mention.

A committee of the Academy consisting of Dr. Arthur Kelman, Dr. Ernest O. Beal, Dr. Henry A. Shannon, and Dr. Herbert E. Speece served on the selection committee for the North Carolina Science Talent Search. Those selected as winners and worthy of honorable mention are listed below. They have been recommended to the college of their choice for scholastic aid or other inducements.

Data is being gathered for consideration of the College Foundation Scholarship Committee for its action when the amount of money available is known.

In an effort to increase the quality and number of participants, a letter was sent to each of the science club sponsors in North Carolina in addition to the usual channels of information such as science fairs and Junior Science Symposium.

Winners

Fairchild, William Royal,* 820 Westover Ave., Winston-Salem
Harmon, Samuel Douglas,* 236 8th St., N. W., Hickory
Kling, Charles Lee,* 1309 N. Market St., Washington, N. C.
Johnson, Betty Ruth, 2702 Fairview Road, Raleigh

Honorable Mention

Blackwell, Richard Edward, 411 Carey Street, High Point
Theys, John Cornelius, Jr., Route 4, Raleigh
Schuitmaker, Harold George, 230 Pennsylvania Ave., Winston-Salem
Ziglar, Paul, 1508 Powell Drive, Raleigh
Spencer, Dennis Odell, 1048 17th Ave., N. W., Hickory

*National Honorable Mention

Report of the Academy Conference

The 1960 meeting of the Academy Conference was held during the AAAS meetings in New York on December 27, 1960. John A. Yarbrough and Charles M. Allen represented the North Carolina Academy of Science. The morning session was concerned with routine reports by various committees and news from academies about their activities during the year 1960. During the afternoon session, President-elect Robert C. Miller of the California Academy of Science presided over a panel discussion concerned with various ways that academies of science have been utilizing their National Science Foundation grants. There is much interest in developing projects that will utilize these opportunities to the fullest extent. There was general feeling that the grants made by the National Science Foundation have been of real value in stimulating Academy interest.

The usual Academy Conference dinner session by courtesy of AAAS was held in the early evening. Dr. J. G. Arnold, Jr., of the Louisiana Academy of Science gave the presidential address.

On the preceding afternoon, December 26, the Academy Conference sponsored a special session relating to junior academies. Mr. E. M. Gurr of the Arizona Academy of Science arranged and presided over this meeting.

Report on Council Meetings of AAAS

Two meetings of the Council of the AAAS were held in New York on December 27 and 30, 1960. It is of particular interest to note that Dr. Paul M. Gross of Duke University has been named as president-elect of AAAS for 1961. Dr. Alan T. Waterman and Dr. Don K. Price were elected to the Board of Directors for four-year terms. Dr. Henry Eyring was selected to complete a one-year term on the Board of Directors.

Other than routine business and minor changes made in the constitution and by-laws, the major interest in the meeting concerned the activities of a special committee studying future efforts of the Council to participate more fully in association affairs. Other matters of interest were reports of committees on science in the promotion of human welfare and discussion of the work of new special committees on the study of specific topics of interest to the Council.

JOHN A. YARBROUGH, *Representative*

Report of the Resolutions Committee

For the membership of the North Carolina Academy of Science we express our appreciation:

1. To Dr. John T. Caldwell, chancellor, and to the faculty and students of North Carolina

State College for their gracious hospitality at the fifty-eighth annual meeting of the Academy.

2. To Dr. Richard H. Loeppert and members of his committee for their efficiency in planning and carying out the many details necessary in arranging the meeting.
3. To the general officers and the sectional chairmen of the Academy for their foresight in preparing a program of interest and value to all members.
4. To the program participants for the diligence with which they have prepared their papers and the clarity with which they have presented them.

PAULINE LONGEST, *Chairman*

The above resolution was unanimously adopted by the Academy.

Treasurer's Report

Financial Statement as of July 10, 1961

Receipts—Academy proper	
Checking account adjusted balance on July 5, 1960	$1,129.88
Dues from memberships, regular	1,592.50
Dues from patron members	120.00
Dues from sustaining members	500.00
Collegiate Academy gifts	125.00
Collegiate Academy funds deposited	11.07
North Carolina Psychological Association for programs	7.50
Exhibit fees, commercial	50.00
Budget allowance for secretarial work from Junior Science Symposium	90.00
From banquet tickets, annual meeting	.50
Total Receipts, Academy proper	$3,626.45
Receipts, Science Fair Program	
Entry fees from seven districts	$1,325.00
Medical Society of North Carolina	100.00
Medical Auxiliary, Medical Society of North Carolina	50.00
American Chemical Society, North Carolina Section	75.00
North Carolina Society of Bacteriologists	50.00
North Carolina Dental Society	100.00
Carolina Power and Light Company	175.00
Southern School Supply Company	100.43
J. D. Taylor for Denoyer-Geppert Company	50.00
Carolina Biological Supply Company	50.00
Cardinal Products, Inc.	100.00
North Carolina Pharmaceutical Association	100.00
Refunds from expenses of National Fair party	59.05
Total Receipts, Science Fairs	$2,334.48
Total Receipts for Academy and Science Fair Program	$5,960.93
Expenditures—Academy proper	
Collegiate Academy, awards and *Cancas*	$ 241.70
Research grants made	1,100.00
Postage, stationery, office supplies	323.13
Expenses for annual meeting, 1961	191.57
Printing	146.10
Secretarial aid	92.25
Secretary-Treasurer's honorarium, remainder of 1959–60	68.65
Secretary-Treasurer's honorarium, 1960–61	150.00
Secretary-Treasurer's travel	39.29
Telephone, long distance	40.09
Expense representation to AAAS	125.00
Elisha Mitchell Journal contributions	852.00
Bond, three years for Secretary-Treasurer	12.50
Service charge, bank	.85
Office secretary, for NSF and Academy work, nine months	751.50
Meals for teachers in NSF institutes	1,187.10
Total Expenditures Academy proper	$5,321.73
Expenditures, Science Fairs	
Announcement brochure, 1961 fairs	$ 100.43
Entrance fee, National Fair	175.00
To Duke University for expense of State Fair	200.00
Prizes at State Fair	220.00
Expense for National Fair party, Kansas City, five persons	1,021.00
Luncheon and travel for planning session, 1962 fairs	71.26
Miscellaneous expense	126.14
Total Expenditures for Fairs	$1,913.83
Total Expenditures for Academy and Fairs	$7,235.56
Bank balance as of June 30, 1961	$ 540.26
Plus deposits to July 10, 1961	2,081.00
Total	$2,621.26
Less outstanding checks, not returned by July 10, 1961	1,799.15
Adjusted bank balance as of July 10, 1961	822.11

Checking account, adjusted as above	822.11
Raleigh Savings and Loan account as of July 10, 1961	1,451.46
Total Assets	$2,273.57

Note: Because the NSF funds we have handled through 1960–61 are still being utilized at the date of July 10, 1961, our total assets cannot be shown. Indirect costs on the grant, as yet not precisely determined, will revert to the Academy.

On the twenty-third day of September, 1961, we, the undersigned examined this account and found it to be in order.

Robert L. Rabb, Chairman
Nash N. Winstead
Henry S. Brown

Memorials were read to the following former members: B. B. Fulton, Walter M. Kulash, E. M. Walker, W. E. Speas, and George G. Ramsey. The memorials were approved by a rising vote of the members present.

Bentley B. Fulton

Bentley B. Fulton, Professor Emeritus in Entomology at North Carolina State College, died at his home December 8, 1960, after a long illness. He was born August 29, 1889 at Newark, Ohio. He received his B. A. degree from Ohio State University in 1912, his M. S. degree from the University of Chicago in 1916, and his Ph. D. from Iowa State College in 1926. He became a member of the North Carolina Academy of Science soon after he joined the State College faculty in 1928 and presented many interesting papers in the meetings of this Academy through the years. He was made a life member in 1955. He was also a member of the Entomological Society of America, the American Association for the Advancement of Science, the Ecological Society of America, the Society for the Study of Evolution, and Sigma Xi.

Dr. Fulton served as assistant entomologist in the New York Agricultural Experiment Station at Geneva from 1912–1919; associate entomologist in the Oregon Agricultural Experiment Station from 1919–1924; assistant entomologist at Iowa State College and Experiment Station from 1924–1928; and professor of Entomology at North Carolina State College since 1928. He retired in 1954.

Dr. Fulton won world-wide recognition as an artist of entomological subjects. Several of his drawings have been used many times in textbooks, as well as various experiment station publications. He was a keen observer and an enthusiastic researcher. He pioneered in the work of distinguishing species of crickets by their songs in addition to the morphological characters. He was a truly great biologist, but due to his quiet, unassuming attitude, he was most appreciated by those who were closely associated with him. He and this speaker were companions on many collecting trips throughout North Carolina and in the earlier days were companions on several camping trips, in some cases with our families. Possibly no one was in a better position to appreciate his keenness of observation, his objectivity, and his intellectual capacity.

He is survived by his widow, two daughters, and four grandchildren.

T. B. MITCHELL

Walter M. Kulash

Walter M. Kulash, Research Professor of Entomology, North Carolina State College, Raleigh, died after a brief illness at Raleigh, North Carolina on September 18, 1960, at the age of 48. Dr. Kulash was born at Haydenville, Massachusetts, on January 8, 1912. He attended the University of Massachusetts, where he received his B. S., M. S. and Ph. D. degrees, the last in 1942. In that year Dr. Kulash joined the staff at North Carolina State College, where he was very active in teaching and in applied entomology, especially in connection with wireworms and insect pests that attack tobacco, corn, cotton, and other farm crops.

Dr. Kulash held membership in many professional and scientific societies, among them the Entomological Society of America, the North Carolina Academy of Science, Sigma Xi, the Society of Systematic Zoologists, the Coleopterists' Society, the American Association of University Professors, and the North Carolina Entomology Society.

Dr. Kulash is survived by his widow, Mary Gobush Kulash; a daughter, Sister Mary Stephanie, of Mount St. Mary's Novitiate, in Newburg, N. Y.; five sons, Walter M., Jr., Dameon J., Joseph A., John A., and Francis M., all of the home.

Dr. Kulash was a former president of the North Carolina Entomology Society, a former president of the State College Chapter of the American Association of University Professors, former secretary of the Zoology Section of the North Carolina Academy of Science, and

former president of the North Carolina Catholic Laymen's Association.

Dr. Kulash had a rich and many-sided life. He was a student of philosophy and languages. He was deeply interested in the development of the students, the department, the college, and the community. He was author or co-author of more than 67 technical articles, publications, and scientific manuals. He exemplified selfless dedication to his church, to his family, and to his work.

C. F. SMITH

E. M. Walker

Ernest Marshall Walker was born May 30, 1911, and died October 30, 1960, of cancer.

He was an honored and beloved member of Campbell College faculty from 1946 to 1960. His students, in dedicating the Annual to him, said of him:

Professor Walker has demonstrated the knack of organizing his life for a maximum contribution. As a teacher of biology, he has stimulated enthusiasm for his subject. As an ordained minister, he had helped in the work of many churches and shared the benefits of his experience with younger brothers on the campus. As a competent business man, he has demonstrated sound practices and good judgment. As a person, he has accumulated a host of friends.

He was a member of the North Carolina Academy of Science for many years. He was a member, and secretary for several years, of the North Carolina Shell Club.

His friendliness will always be remembered and missed by all of those who knew him.

NANCY BLAIR ELIASON

William Eugene Speas

William Eugene Speas was born in Yadkin County on December 1, 1885, and died at his home in Winston-Salem on January 24, 1961.

After receiving his Bachelor of Arts degree from Wake Forest College in 1907, he completed his work at The Johns Hokpins University for the Master of Arts in 1913, and at Cornell for the Doctor of Philosophy in 1927. Dr. Speas taught physics in Clemson College from 1913 to 1920. He joined the Wake Forest College faculty in 1920 and taught in the Department of Physics until his retirement in 1959, serving for a quarter of a century as chairman of his department.

He loved beauty, whether he found it in the vocal and instrumental music of the members of his devoted and talented family, in the magnificent "mums" which he raised in his flower garden, or in what he felt to be the created order of God's universe.

He combined to a remarkable degree the happy faculty of scholar and teacher. While serving with distinction as President of the North Carolina Academy of Science in 1938, he was teaching physics to both beginning and senior students with a contagious enthusiasm and genuine joy that continued through the last class he met.

The effects of this enthusiasm and joy can best be seen in Dr. Speas' students. Upon the death of this beloved professor one of his former students wrote to the college newspaper.

The most interesting days in physics were those days when Dr. Speas brought his homemade apparatus to class and demonstrated the truth of what the textbooks said. One demonstration involved a coffee can lid and a little springloaded gun with a steel ball for a bullet. The demonstration proved that falling objects tend to fall at the same rate of speed, regardless of what horizontal forces might be at work.

Actually, it was a simple sort of demonstration. But Dr. Speas liked it. On the day before he showed it to us he told us to be sure and be in class.

"It's a beautiful experiment," he said.

From that day on I had more respect not only for Dr. Speas, but for the subject he was teaching. Physics—the patterns, the workings of the world around him—was "beautiful" to this man.

The English professor with his volume of Shakespeare, the musician with his fine instrument—they had nothing on Dr. Speas. He, too, had found beauty in his work.

And I realized that morning that if physics was not "beautiful" to me, then I was missing out.

Many students at registration have said "My father, who had Dr. Speas for physics, told me to be sure and get in his class or my education would not be complete."

This effect upon his many students is the greatest tribute to Dr. Speas, a scholarly and enthusiastic teacher, a devoted husband and father, and a wholesome Christian neighbor.

T. J. TURNER

George Garfield Ramsey

George Garfield Ramsey died November 9, 1960, at the age of eighty years.

Dr. Ramsey received the A. B. degree and the M. S. degree from the Greensburgh Seminary, Greensburgh, Pennsylvania, and the M. A. and Ph. D. degrees from Columbia University. He was chairman of the Chemistry department at Catawba College until his retirement after 27 years of service. He made

significant contributions to knowledge of temperature coefficients of enzymatic activity and of thermal destruction of trypsin. Among his professional affiliations were The American Chemical Society, Sigma Xi, and the North Carolina Academy of Science.

He was a man of many virtues, but of such reserve and modesty that few persons knew him sufficiently well to appreciate him fully.

To his colleagues, he was a true scholar and a meticulous research worker. Though by nature a self-sufficient and retiring person, his unfailing consideration of the rights of others and the reliability of his statements made his presence welcome and valued in faculty business.

To his students, he was an exacting taskmaster. Any workmanship of quality less than good was not acceptable, and "fair" was not worthy of discussion. The interested student found in him a teacher who was always ready to provide opportunity for independent work, the encouragement for study beyond the assignment and the challenge to reach the very limit of his capacity for excellence.

Dr. Ramsey is survived by one close relative. But scores of his former students, now active in various fields of science, cherish his memory and honor his philosophy that Truth is to be sought for the adventure, not as a destination.

DANIEL E. KIRK

Report of the Nominating Committee

The committee, consisting of John F. Dashiell, John W. Lasley, and Reinard Harkema submitted the following nominations:

President: Paul J. Kramer, Duke University

Vice-President: Charles E. Jenner, University of North Carolina

Member of Executive Committee: Victor M. Greulach, University of North Carolina

Member of Research Grants Committee: F. J. Vernberg, Duke University

Secretary-Treasurer (for three-year term): John A. Yarbrough, Meredith College

Editor (for three-year term): Reinard Harkema, North Carolina State College

The above nominees were unanimously elected by the Academy. This concluded the business and the meeting was adjourned.

Approximately 125 members of the Academy, guests, and friends enjoyed the Annual Dinner in Leazer Hall at 6:15 p.m. Vice-President E. G. Purdom presided at the dinner and the brief session which followed. The 1960 Poteat Award, presented annually by Phipps & Bird, Inc., of Richmond, Virginia, was formally presented to Dr. Wesley O. Doggett of North Carolina State College for his paper in 1960 in the Mathematics section entitled An Infinite Set of Elementary Functions. Because of the large number of papers presented in the Collegiate Academy session, two awards were made again this year, one in Biological Science and one in Physical Science. W. Keith Baucom and Kenneth M. Lewis of North Carolina State College were the joint authors of the winning paper in Biological Science, and Arlan Mantz of Catawba presented the winning paper in Physical Science.

Following the banquet, the members and friends adjourned to the Lecture Room in Williams Hall, where President Jensen presented his presidential address, The Scientific Quest for Significance. Vice-President Purdom presided and presented Dr. Jensen to the audience. Following the lecture all members and guests were entertained at a very pleasant social hour through the courtesy of North Carolina State College in the McKimmon Room of Williams Hall.

The following officers were elected for the respective sections:

Biochemistry and Physiology: Chairman, C. Artom; Secretary, D. S. Grosch

Botany: Chairman, B. W. Smith; Secretary, Christine Wilton

Geoscience: Chairman, William J. Furbish; Secretary, S. Duncan Heron, Jr.

Mathematics: Chairman, John B. Davis; Secretary, I. C. Gentry

Physics: Chairman, J. W. Straley; Secretary, Anna J. Reardon

Zoology: Chairman, A. D. Shaftesbury; Secretary, F. J. Vernberg

The following constitute the personnel of the standing committees:

Executive: Paul J. Kramer, C. E. Jenner, John A. Yarbrough, Reinard Harkema, Jesse P. Tyndall, Mrs. Pauline Longest, and Victor M. Greulach

Research Grants: John M. Parker, III, M. K. Berkut, V. M. Cutter, Jr., and F. J. Vernberg

Other committees will be appointed in the early fall of 1961 for the school year 1961–62.

Program

The following papers were presented during the meetings at Raleigh. Those marked with an asterisk are abstracted in these proceedings:

GENERAL SESSION

Health physics. Dr. Newton Underwood, Department of Physics, N. C. State College

The American gray squirrel in Great Britain. Dr. F. S. Barkalow, Jr., Department of Zoology, N. C. State College

Recent progress in low temperature physics. Dr. Horst Meyer, Department of Physics, Duke University

Sex reversal in plants? Dr. Frederick A. Wolf, Department of Botany, Duke University

COLLEGIATE ACADEMY

Biological Science

Effects of hybridization on longevity of *Drosophila* adults. W. Keith Baucom and Kenneth M. Lewis, North Carolina State College

Context effects on cross-modality equations of sensory magnitude. Ann Howell Hardy, Woman's College, U. N. C.

Response of pre-puberal ovarian tissue to hormone stimulation. Talmadge T. Brown, Jr., North Carolina State College

An investigation of parasitism in *Cambarincola vitrea* (family Branchiobdellidae) on their crayfish host. Clarence E. Styron, Jr., Davidson College

The effect of trichinosis on the vitality of the white mouse. Roxanna L. Neikirk, Catawba College

A study of population growth in *Carteria* sp. cultures as influenced by *Nitzschia closterium*. James R. Wheatley, Jr., East Carolina College

Studies on the ecology of the giant mosquito, *Toxorhynchites rutilus*. Mrs. Anne McCrary, U. N. C.

Physical Science

Glass fiber and paraffin-coated papers in the isolation of fat-soluble compounds. James O. Rice, A. & T. College

Ultraviolet absorption spectra of compounds related to cholesterol. John C. Holley, A. & T. College

The optical properties of several types of hardware cloth used as achromatic filters in photometric determinations. Arlan Mantz, Catawba College

Electrophoretic separation of proteinases of *Lumbricus terrestris*. Robert C. Costello, U. N. C.

The achromatic contrast functions of six observers using a photometric simultaneous contrast comparator. Doris Steppe and David Saleeby, Catawba College

A comparative study of tissue proteins by color electrophoresis. Walter J. Harrison, A. & T. College

BIOCHEMISTRY AND PHYSIOLOGY SECTION

*Physiologic factors which limit the minimal utilizable oxygen for rats in a microenvironment. F. G. Hall, Duke University

*Chemiluminescence caused by milk xanthine oxidase. Lorrance Greenlee and Irwin Fridovich, Duke University

*Effects of exogenous female hormones in the male reproductive organ. N. Pati and C. F. Thompson, Jr., A. & T. College

*Effects of crude lipid extracts of hypothalmus on reproduction. Hugh A. Poston and J. A. Santolucito, N. C. State College

*The mechanism of radioactive zinc uptake by *Ulva lactuca*. John Gutknecht, U. N. C.

*Methionine in cholesterol synthesis. Cecile H. Edwards, Evelyn L. Gladsden and Gerald A. Edwards, A. & T. College

*Uptake of methionine-2C^{14} and methionine-methyl-C^{14} by tissues of hepatoligated rats. Gerald A. Edwards, Cecile H. Edwards and Evelyn L. Gladsden, A. & T. College

*Physico-chemical properties of deoxypentose nucleohistones. J. L. Irvin and C. McClure, U. N. C.

*Clarification of the foreign bone-marrow reaction in irradiated mice through *in vitro* enzyme studies. Gladys W. Royal, George C. Royal, Jr., and Evelyn P. Mills, A. & T. College

BOTANY, SECTION A

*A preliminary floristic and vegetational study of the Toxaway River Gorge, Transylvania County, North Carolina. Arthur W. Cooper, N. C. State College

Distribution of ponderosa pine on the northern Coconino Plateau, Arizona. Patricia Rand, Duke University

*Patterns of variation in eastern erythroniums. Clifford R. Parks, N. C. State College

*Determining tree root extension with radioactive tracers: a progress report. M. L. McCormack and F. W. Woods, Duke University

*Using towers for teaching microclimatic stratification. F. W. Woods and M. L. McCormack, Duke University

*The significance of interspecific fertility in the evolution of pathogenicity in the genus

Helminthosporium. R. R. Nelson, N. C. State College

*The pathogenicity of twelve known and nine unidentified gramineous species of *Helminthosporium* to twenty-three genera of the Gramineae. R. R. Nelson and D. M. Kline, N. C. State College

*The role of heat in predisposing peanuts to infection by *Diplodia gossypina* Cke. J. M. McGuire and W. E. Cooper, N. C. State College

*Numerical unbalance of chromosomes in synthetic pentaploids of tobacco and cotton. S. J. Yang and F. A. Bernardo, N. C. State College

BOTANY, SECTION B

*Some cultural aspects of two species of the genus *Archangium* (Myxobacteriales). James C. McDonald, Wake Forest College, and John E. Peterson, University of Missouri

Effect of a current on mineral uptake and respiration by a green alga. L. A. Whitford, N. C. State College, and G. J. Schumacher, State University of New York

Influences of gibberellic acid on several species of algae. Won K. Kim and Victor Greulach, U. N. C.

Some evolutionary aspects of *Sphagnum*. H. L. Blomquist, Duke University

*Relationship of the vascular tissues in the major and minor veins of foliage leaves. Brenda Slade, University of Otago, New Zealand

Investigation into foliar anatomy and ontogeny of certain species of *Ilex* L. R. P. Ashworth, Clemson College

*Blade tissue studies of Sonoran desert shrubs. Jane Philpott, Duke University

GEOSCIENCE SECTION

*Aspects of negative crystal growth in quartz crystals from the eastern Piedmont of North Carolina. William J. Furbish, Duke University

*Structural geology of the Hanging Rock area, Stokes County, North Carolina. Barry A. Centini, N. C. State College

*Role of clay mineralogical analysis in soil genesis investigations. L. A. Nelson and R. J. McCracken, N. C. State College

*Geology of pryophyllite deposits near Glendon, Moore County, North Carolina. James F. Conley, N. C. Division of Mineral Resources, Raleigh

*What is the future of the Geology Section in the N. C. Academy of Science? Stephen G. Conrad, Division of Mineral Resources, Raleigh

PHYSICS SECTION

Invited Papers on the Teaching of Physics

A demonstration of new teaching apparatus in use at Chapel Hill. Joseph Straley, U. N. C.

*Textbooks in use in North Carolina colleges and universities. Marshall Helms, East Carolina College

Contributed Papers

*The in-service institute as an aid to the high school physics teacher. Sherwood Githens, U. S. Army Research Office (Durham)

*A report on the recent mathematics-physics academic year institute curriculum conference at Washington University. W. E. Haisley, U. N. C.

*A new method for determining the true count rate from a nuclear detector. W. O. Doggett, N. C. State College

*Radiation reaction in a gravitational field. R. W. Brehme, Wake Forest College

*Cosmological spaces admitting geodesic correspondence. B. E. Leonard and W. R. Davis, N. C. State College

*A generalized variational method for solving differential equations of physics. A. L. Mowery and R. L. Murray, N. C. State College

*Analytical studies in neutron slowing and diffusion. Donald Sells and R. L. Murray, N. C. State College

*A laboratory experiment using neutron threshold detectors. David H. Martin, N. C. State College

ZOOLOGY, SECTION A

The ecology of parasitism in the raccoon on Cape Island, South Carolina. Reinard Harkema and Grover C. Miller, N. C. State College

*A new strigeoid trematode (Diplostomatidae: Alariinae) from Florida raccoons. Reinard Harkema and Grover C. Miller, N. C. State College

*Preliminary studies on the life history of *Heterobilharzia americana* Price, 1929. Nathan D. Greene, N. C. State College

Patterns in biological nomenclature from savage to scientist. A. L. Pickens, Charlotte

*Microstratification of the Tomocerinae (*Collembola*) in a beech-maple forest of eastern North Carolina. Clifford B. Knight, East Carolina College

*Pattern of emergence in the dragonfly *Tetragoneuria cynosura*. Paul E. Lutz, U. N. C.

*Types of induced modifications in the pattern of oviposition of the wasp *Habrobracon*. D. S. Grosch, N. C. State College

*Regeneration of the intestine of *Ambystoma* larvae. Marilyn S. Kerr, Duke University

*Germinal localization and morphogenesis of midgut derivatives in the salamander, *Ambystoma punctatum*. Thomas M. Harris, U. N. C.

*The effects of removal of portions of kidneys on cellular proliferation in tubular epithelia of remaining kidney tissue. Charles E. McCreight, Bowman Gray School of Medicine

*Maintenance of the unhydrated state of egg jelly in the *Rana pipiens* reproductive tract. Thomas W. Lee, Duke University

*Differentiation of *Hydra* interstitial cells in culture. Maximo Cerame-Vivas and C. G. Bookhout, Duke University

ZOOLOGY, SECTION B

*Siamese pigeons. R. W. Prichard and Torguan Tan, Bowman Gray School of Medicine

A study of the golden mouse, *Peromyscus nuttalli aureolus*. James H. Eads, Jr., Meredith College

*Some fishes from the area of Chicoteague Bay, Virginia. L. M. Outten, Mars Hill College

*Observations on the spawning coloration and behavior of some cyprinid fishes. L. M. Outten, Mars Hill College

Factors in the conditioning of aggressive behavior in the Siamese fighting fish, *Betta splendens*. James H. Horton, Jr., and Robert R. Haubrich, East Carolina College

*Ammonia and urea excretion in some paedomorphic urodeles. Richard C. Bruce, Duke University Marine Laboratory

Ecology of the terrestrial isopod *Armadillidium vulgare*: food, enemies, weather. Oscar H. Paris, U. N. C.

*Random observations of fresh-water invertebrates. Tom Daggy, Davidson College

The effect of osmotic stress on the body fluid in the ascidian *Styela plicata*. Mary W. Pinschmidt, Duke University Marine Laboratory

*The influence of thermal and osmotic stress on the body fluid on the ciliary activity of gills of lamellibranchs. F. J. Vernberg, David E. Schneider, and Carl Schlieper, Duke University Marine Laboratory

*Larval distribution of three species of *Balanomorpha* in relation to some physicochemical factors. Murad.-B. M. Mohammad, Duke University Marine Laboratory

Abstracts

PHYSIOLOGIC FACTORS WHICH LIMIT THE MINIMAL UTILIZABLE OXYGEN FOR RATS IN A MICROENVIRONMENT. *F. G. Hall.* Hypoxia and body temperature operate reciprocally in determining the utilization of oxygen of rats in a closed environment.

Body temperature during hypoxia can be maintained at normal levels when environmental temperatures are between 32°C. and 33°C.

Critical oxygen tensions of rats in a closed microenvironment are influenced by body temperatures. Lower body temperatures permit lower critical oxygen tensions.

Acclimatization to hypoxia has little effect upon minimal critical oxygen tension and any significant affect is adverse. Hemoglobin affinity for oxygen appears to be more important than the quantity of hemoglobin present.

CHEMILUMINESCENCE CAUSED BY MILK XANTHINE OXIDASE. *Lorrance Greenlee and Irwin Fridovich.* Aminophthalhydrazide (luminol) and dimethyl biacridyllium nitrate (lucigenin) are known to emit light upon reaction with hydrogen peroxide. It has been reported (J. R. Totter, et al. 1960. J. Biol. Chem. **235**: 238) that xanthine oxidase, when acting on its substrates in the presence of luminol or lucigenin, will cause emission of light. The intensity of this enzymically induced luminescence was thousands of times too intense to be accounted for in terms of the hydrogen peroxide produced by the enzyme. Certain of the peculiarities of the kinetics of light emission were interpreted as indicating the necessity for the simultaneous binding of two molecules of substrate on the enzyme. Since it had been demonstrated in this laboratory earlier that xanthine oxidase, when acting on its substrates in the presence of oxygen, generates reactive radicals which were detectable by chemical means, it appeared likely that chemiluminescence was a consequence of the reaction of these enzyme-generated radicals with luminol or lucigenin. A study of this enzyme-induced luminescence was therefore undertaken.

Proteins such as bovine serum albumin strongly enhance the intensity of light emission, probably by binding lucigenin and thereby increasing the efficiency of conversion of excitation energy to light. In the presence of optimal levels of bovine serum albumin, many of the kinetic peculiarities of the system vanish,

as does the necessity for postulating simultaneous binding of two substrate molecules. The action of xanthine oxidase on lucigenin appears to be mediated by the reactive superoxide radicals ($O_2^{-\cdot}$) generated by the enzyme. Rabbit liver aldehyde oxidase, which has been found to generate these radicals, also induces this chemiluminescence, the study of which may shed light both on the mechanism of action of these enzymes and on the luminescent reaction itself.

EFFECTS OF EXOGENOUS FEMALE HORMONES IN MALE REPRODUCTIVE ORGAN. *N. Pati and C. F. Thompson, Jr.* Sixteen male albino rats of the Holtzman strain, each weighing approximately 130 ± grams and from 80 to 90 days old, were randomly divided into four groups. Each animal in each of the three treated groups was given separate subcutaneous injections of stilbesterol and progesterone at the rate of .5 mg. of each per day and a combination of stilbesterol and progesterone at the rate of .25 mg. of each per day consecutively for three weeks. The control group was receiving physiological saline solution at the rate of 1 cc. per day per animal. At the end of three weeks two animals from each group were killed and the testes removed for histological study. The size of the testes was increased in animals treated with progesterone in comparison with the others as well as the control. Histological study revealed mitosis of the germ cells, cells of Sertoli, and dense connective tissue. Tail of the spermatozoa was filiform, and a large number of normal spermatozoa was observed.

Histological sections of testes treated with estrogen showed a noticeable change in abnormalities characterized by the absence of spermatozoa, abnormal meiosis and hypertrophied condition of the spermatid.

Observation in the sections of testes treated with combination of estrogen and progesterone revealed abnormal meiosis as well as increase in the size of spermatid. Cells of Sertoli were much more flattened and conical rather than cornified. Individual nuclei showed an increase in chromatin granules. Cytoplasmic material was very faint and evidence of rapid abnormal meiosis was indicated by the presence of many nuclei in one cell membrane, a condition which is postulated to be a nuclear cyst.

EFFECTS OF CRUDE LIPID EXTRACTS OF HYPOTHALAMUS ON REPRODUCTION. *H. A. Poston, L. C. Ulberg, and J. A. Santolucito.* Twenty-five mg. of lipid extracted from bovine hypothalamus and administered in cottonseed oil (0.5 ml.) as a single, subcutaneous injection resulted in depression of body, reproductive tract, and pituitary gland weights in male and female rats or mice. For example, in one trial with seven female mice (21–23 days) the following percentage weight reductions (96 hrs. post-injection) were obtained: body, –40; ovaries, –10; uterus, –10; pituitary, –51; adrenals, –15. Similarly, for 14 male mice: body, –13; testes, –36; ventral prostate, –28; seminal vesicles, –38; pituitary, –17; adrenals –8. The above weight depressions, though slight, were consistent. Of interest was whether this effect could interfere with reproductive performance. Treated animals received daily subcutaneous injections of 15 mg. of lipid extract beginning on the day of finding sperms in the vaginal smear, and were autopsied 24 hours after final injection. The control animals received daily subcutaneous injections of cottonseed oil. Ten animals autopsied on Day 5, when compared with their controls, indicated no apparent influences on ovulation rate or on embryonic development to time of implantation. Embryos observed at this time were in the inner-cell-mass stage. However, by Day 12 of gestation, the average data for 19 treated animals showed 4.0 more corpora lutea than normal fetuses, while their controls exhibited a discrepancy of 1.5 for this measurement of loss of potential young. Average numbers of corpora lutea were similar in treated and control animals. In another series of animals which were allowed to go to term, daily subcutaneous injections were administered for the first 19 days of gestation. The average number of live young born for this group was 7.4 while their controls littered an average of 11.4 It was interesting to note that postnatal loss of the treated group prior to weaning was approximately twice the magnitude of that in the controls. The differences in the averages between the treated and control animals were largely due to rather marked effects in a few animals, rather than a slight effect on all animals.

THE MECHANISM OF RADIOACTIVE ZINC UPTAKE IN *Ulva lactuca*. *John Gutknecht.* The effects of metabolism, *p*H, carrier ions, and temperature upon the uptake and accumulation of Zn^{65} by *Ulva lactuca* from sea water in the light and the dark were investigated. The pattern of uptake in relation to light and temperature in unbuffered sea water was what would be expected assuming zinc absorption to be a biological accumulation process proportional to the rate of photosynthesis as has been previously reported. However, uptake rate and

level of accumulation were found to be strongly *p*H dependent and to be affected by the concentration of zinc in the media. Freshly killed *Ulva* absorbed more tracer than live material. These findings suggest that the physical processes of absorption and ionic exchange are primarily responsible for Zn^{65} uptake. The relationship between photosynthesis and zinc adsorption is a secondary effect related to surface/volume ratio and *p*H.

METHIONINE IN CHOLESTEROL SYNTHESIS. *Cecile H. Edwards, Evelyn L. Gadsden, and Gerald A. Edwards.* Digitonin is widely used in analytical procedures for the quantitation of cholesterol in animal tissues. We have employed this compound and the glycoside tomatine to study the incorporation of methionine, labeled in the methyl or *alpha* carbon, into tissue cholesterol.

Cholesterol in alcohol:acetone:ether extracts of the liver of adult male rats fed either methionine-methyl-C^{14} or methionine-2-C^{14} was precipitated by the addition of digitonin or tomatine.

The specific activities in c/m/mg. of the cholesterol and cholesterol ester digitonides from the liver of a rat fed methionine-methyl-C^{14} were 99 and 419 respectively; coprostanol digitonide from feces contained 546 c/m/mg. Cholesterol and cholesterol ester tomatinides contained 510 and 546 c/m/mg. whereas the radiocarbon in these compounds isolated after feeding methionine-2-C^{14} was only slightly above background.

The melting points and ultra violet and infra-red absorption curves of tomatinides prepared from pure cholesterol and rat liver were identical.

These data suggested that the methyl carbon of methionine, but not its *alpha* carbon, is used for cholesterol synthesis.

(Supported in part by Grant A-1464 from the National Institutes of Health, Public Health Service).

UPTAKE OF METHIONINE-2-C^{14} AND METHIONINE-METHYL-C^{14} BY TISSUES OF HEPATOLIGATED RATS. *Gerald A. Edwards, Cecile H. Edwards, and Evelyn L. Gadsden.* It is well known that many important functions in the metabolism of nutrients are performed by the liver. Among these is the process of demethylation in which methionine is converted to homocysteine. Adult male rats were hepatoligated by a procedure developed in our laboratory. Experimental animals so treated and sham-operated controls were sacrificed 30 minutes after the injection of methionine-2-C^{14} or methionine-methyl-C^{14}.

When significantly less methionine was received by the liver due to diversion of the portal blood to the vena cava, strikingly smaller concentrations were incorporated into tissues.

The utilization of methionine for tissue protein synthesis was markedly decreased, presumably due to interference with the demethylation of this amino acid.

(Supported in part by Grant A-1464 from the National Institutes of Health, Public Health Service).

PHYSICAL CHEMICAL PROPERTIES OF DEOXYPENTOSE NUCLEOHISTONES. *J. Logan Irvin and C. McClure.* Unfractionated histones were prepared from deoxypentose nucleoprotein of calf thymus and rat liver by extraction with 0.2 N and 0.5 N HCl, respectively. Arginine- and lysine-rich fractions were obtained from thymus by ethanol fractionation at −10°C. Starch gel electrophoresis (*p*H, 4.76; HOAc-NaOAc buffer; ionic strength, 0.02) of thymus unfractionated, arginine, and lysine histones reveals 8, 6, and 5 components, respectively. Rat-liver histones are composed of 7 subfractions. Ultraviolet spectroscopy reveals an absorbancy maximum at 276 mμ with 276/260 = 1.8 for thymus histones and an absorbancy maximum at 273 mμ with 273/260 = 1.1 for rat-liver histones. Electrophoretic data strongly suggest species and/or tissue specificity. The hypsochromic shift in the ultraviolet suggests that this difference may reside, in part, in a greater per cent covalent character in the secondary and tertiary bonds in the rat-liver protein. These postulates are supported by the concentration of acid required for more complete extraction of rat-liver deoxypentose nucleoprotein.

CLARIFICATION OF THE FOREIGN BONE-MARROW REACTION IN IRRADIATED MICE THROUGH *In Vitro* ENZYME STUDIES. *Gladys W. Royal, George C. Royal, and Evelyn P. Mills.* Liver, lung, spleen, kidney and brain homogenates from normal $(101 \times C_3H)F_1$ male mice 10 to 12 weeks of age were incubated *in vitro* in enzymatic proportions with total-body protein isolates from rats and mice. The rate of oxygen uptake was determined for each of the mixtures. Neither homogenate produced appreciable activity with isologous protein; however, liver and spleen reacted significantly with the heterologous (rat) isolate. Prolonged *in vitro* incubation of from 14 to 16 hours for mouse spleen homogenate with rat bone marrow, prior to injection in x-irradiated mice, produces mor-

tality in the same sample proportion at the *in vivo* foreign bone-marrow reaction, though at an increased rate. Spectrophotometric and electrophoretic comparisons of fresh and incubated rat bone marrow were made. Histological and spectrophotometric studies of tissues from animals experiencing the coupled *in vivo, in vitro* reaction were compared with those having responses to the *in vivo* reaction alone. Implications of the technique in circumventing the *in vivo* response are given.

(Supported by U. S. Atomic Energy Commission Contract No. AT-(40-1) 2399.)

A PRELIMINARY FLORISTIC AND VEGETATIONAL STUDY OF THE TOXAWAY RIVER GORGE, TRANSYLVANIA COUNTY, NORTH CAROLINA. *Arthur W. Cooper.* A preliminary floristic and vegetational survey of the Toxaway River Gorge, Transylvania County, North Carolina, was carried out during mid-summer of 1960. Observations on species distributions and community types were made from the 1,200 feet elevation to 3,000 feet at Toxaway Falls. A total of 304 vascular plant species was recorded in the gorge. Four species of primarily coastal plain distribution were recorded and altitudinal limits were noted for a number of woody species. Four major community types were recognized. A shrub zone occurred along river and stream banks throughout. Scrubby Virginia pine stands were found along gravelly and rocky disturbed sites along the river bottom. A mixed mesophytic forest, with three recognizable phases, occurred on river slopes, coves, and gentle slopes along the river. Oak forest covered the greater part of uplands above the gorge and extreme sites within the gorge. A close parallel seemed to exist between vegetation types noted in the Toxaway Gorge and those described for lower elevations in the Great Smoky Mountains.

PATTERNS OF VARIATION IN EASTERN YELLOW ERYTHRONIUMS. *Clifford R. Parks.* The eastern yellow erythroniums have for some time presented a problem of taxonomic complexity which is further complicated by the great variation found in any one population. The present study, however, has led to some possible explanations of this variation. Twenty years ago Dr. R. M. Harper suggested that the erythroniums growing around Durham, North Carolina, were likely a distinct but unnamed species. This diploid type differs from "typical" *E. americanum* in a number of different characteristics, and it appears to be distinct. The two species of this group essentially limited to Alabama and described by Wolf in 1941 also have been studied. The distinctness of the diploid *E. rostratum* is beyond question, but the tetraploid *E. Harperi* may represent a deep southern race or subspecies of *E. americanum*, or possibly a separate species. The status of the erythroniums of the high, moist habitats of the Southern Appalachians is not yet known for certain. These appear to be more closely related to the piedmont species, but they consistently differ from it in a number of characteristics. It can be speculated from morphological and cytological information that the tetraploid *E. americanum* was produced by an ancient hybridization of *E. rostratum* and the piedmont species, and the high elevation type was formed by a similar, but much more recent, cross between *E. americanum* and the piedmont species. Further work, particularly cytological, needs to be done to determine whether this pattern of reticulate evolution is the means by which speciation has occurred in the genus *Erythronium*.

DETERMINING TREE ROOT EXTENSION WITH RADIOACTIVE TRACERS: A PROGRESS REPORT. *M. L. McCormack and F. W. Woods.* Soil applications of ionizing compounds and the use of appropriate field monitoring techniques should provide a qualitative method of determining lateral extent of roots in slash-pine plantations on the sandhills of North Carolina. To this end, two portable scintillation detectors have been designed for use with portable scalers and recorders. Detection problems include correction for local radioactivity in soil, cosmic radiation, and instrument instability. Instrument sensitivity is essential because of the rapid and extreme dilution of isotopes once they are placed in the environment. This work should contribute to our knowledge of tree roots as related to volume of soil occupied, the extent of root competition, and movement of minerals in soils and plants.

USING TOWERS FOR TEACHING MICROCLIMATIC STRATIFICATION. *F. W. Woods and M. L. McCormack.* A set of laboratory exercises for the study of environment is being developed. Designed for use by all students beginning the study of forest ecology, they require the availability of three towers which can be climbed to operate simple meterological instruments at different heights above the forest floor. The studies require about 24 hours of field work and an equal time for preparing reports; however, adjustments can easily be made to suit the needs of individual instructors.

Single copies of prepared materials will be

available to instructors in biology, botany, and forestry departments upon request.

THE SIGNIFICANCE OF INTERSPECIFIC HYBRIDIZATION IN THE EVOLUTION OF PATHOGENICITY IN GENUS *Helminthosporium*. *R. R. Nelson.* Species of *Cochliobolus* having *Helminthosporium* conidial stages possess complex genetic systems controlling sexual reproduction. In spite of this complexity, crosses between species of *Cochliobolus* and between *Cochliobolus* spp. and species of *Helminthosporium* with no known sexual stage have been successful. Seventy-one different fertile crosses were obtained from 17,838 attempted matings of 174 isolates of 16 species, a frequency of .39 percent. Hybridization between pathogenic species of *Helminthosporium* provides the vehicle for recombination of genes for pathogenicity. Evaluations of hybrid ascospore progenies indicate that certain genes are unique to each of the parental species, in that progenies frequently segregate 1:1:1:1, one-half being pathogenic to one or the other parental host, one-fourth being pathogenic to both hosts, and one-fourth being nonpathogenic to both parental hosts. Certain hybrid ascospore progeny are pathogenic to hosts that are resistant to both parental species, demonstrating that although certain gene systems for pathogenicity are specific for certain hosts, species of plant pathogens possess gene pools for pathogenicity beyond the limits required to attack given hosts. Recombinant classes nonpathogenic to both parental hosts are pathogenic to other hosts, indicating that the specific genes required for pathogenicity to one host may not directly influence the remainder of the gene pool although they may be a part of additional systems. It is concluded that pathogenicity potential in species of *Helminthosporium* represents a complex genetic system and that gene recombination by means of interspecific hybridization is an important evolutionary factor.

PATHOGENICITY OF 12 KNOWN AND 9 UNIDENTIFIED GRAMINEOUS SPECIES OF *Helminthosporium* TO SPECIES IN 22 GENERA OF THE GRAMINEAE. *R. R. Nelson and D. M. Kline.* It is commonly believed that most species of *Helminthosporium* causing leafspots on gramineous hosts are restricted in pathogenicity to species in one or a few genera. In the present studies, the pathogenicity of one isolate each of 12 known and 9 unidentified gramineous species of *Helminthosporium* was determined on 28 species in 22 genera of the *Gramineae*. *H. maydis, H. carbonum, H. sorghicola,* and *H. leersiae,* each previously reported as pathogenic only to one or more species of a single genus, were pathogenic to species in 14, 8, 7, and 17 genera, respectively. *H. victoriae* and *H. oryzae,* each previously reported on species in 2 genera, were pathogenic to species in 6 and 18 genera, respectively. *H. sacchari* and *H. cynodontis,* reported on species in 3 genera, and *H. rostratum,* reported on species in 5 genera, caused leafspot symptoms on species in 4, 3, and 11 genera, respectively. Isolates of *H. turcicum* from corn, sorghum, and Sudangrass were pathogenic to species in 3, 3, and 2 genera, respectively, suggesting differences in pathogenicity among isolates from different hosts. Although *H. sativum* has been reported on species in 30 genera, it has been reported as a foliage pathogen of species in 8 genera. In these studies, *H. sativum* produced distinct leaf lesions on species in 12 genera. All of the 9 unidentified species—2 from barley, 3 from Bermuda grass, and 4 from oats—were pathogenic to species in several genera, ranging from 3 to 17. *H. homomorphus,* a species considered to be saprophytic, did not cause a lesion reaction on any of the species tested. In view of the variation in pathogenicity among isolates of fungus species, the data are not intended to indicate the potential host range of these species, but rather to demonstrate that species of *Helminthosporium* are not host specific and that the description of new species solely on the basis of the reaction of one host is not justified.

THE ROLE OF HEAT IN PREDISPOSING PEANUTS TO INFECTION BY *Diplodia gossypina* CKE. *J. M. McGuire and W. E. Cooper. Diplodia gossypina* Cke. was proved the causal agent of collar rot of peanuts. Consistent infection was obtained, however, only when plants were predisposed with heat prior to inoculation. Peanut stems irradiated with a heat lamp until the surface temperature reached 45°C. usually developed heat lesions and were susceptible to *D. gossypina.* Heat-induced lesions rarely developed and collar rot susceptibility was not appreciably altered when the surface temperature of irradiated stems reached only 40°C., whereas a surface temperature of 50°C. usually killed the stem, although subsequent infection by *D. gossypina* was not evident. Internal stem temperatures averaged 14°C. higher than the average surface temperature after 10 minutes of irradiation.

The apical portions of defoliated three-inch sections of peanut stems were also predisposed

to infection by submersion in hot water. Some sections maintained at an internal stem temperature of 46°C. for 12 minutes and then inoculated became infected although no heat injury was evident. Heat lesions developed on sections partially submerged in water at 49° and 52°C. for 4, 8, and 12 minutes, and inoculated sections became infected. Rate of disease development was proportional to severity of heat treatment.

Heat lesions were histologically evident within 24 hours after the plants were heated to a surface temperature of 45°C. with radiant heat. At this time the fungus was well established in cortical tissue on the irradiated side of stems of inoculated plants. The mycelium was largely intercellular. An increase in the exosmosis of reducing sugars from heated stems indicated an increase in cell permeability. This is suggested as a possible cause of increased susceptibility.

Surface temperatures of peanut stems exposed to sunlight under field conditions were from 5° to 10°C. higher than air temperature, and the internal stem temperatures were probably higher, although this was not determined. Therefore, peanut stems exposed to direct sunlight may be predisposed to infection by *D. gossypina*, especially with air temperatures of 35°C. or higher.

NUMERICAL UNBALANCE OF CHROMOSOMES IN SYNTHETIC PENTAPLOIDS OF TOBACCO AND COTTON. *S. J. Yang and F. A. Bernardo.* Amphiploid *Nicotiana* and *Gossypium* were synthesized, and their segregations upon crossing with a cultivated tester have been investigated extensively in the cytogenetics laboratory at North Carolina State College. It was in view of the results from these works that unbalance in chromosome numbers was suspected in the derivatives of artificial amphiploids. The present study confirms the occurrence of various somatic chromosome numbers differing from the expectations in pentaploid materials. Pentaploids of *Nicotiana* were obtained by backcrossing the synthetic hexaploids *N. tabacum-otophora* and *N. tabacum-tomentosiformis* to their tetraploid parent *N. tabacum*. The chromosome number of each plant was determined by assessing a uniform count from five or more cells of root-tip tissue. Within-plant variation was found only occasionally. Among the 69 plants examined, less than half (31 plants) had the expected 60 chromosomes. There were twenty-five plants with 59, five with 56, 57 or 58, and eight with 61 or 62 chromosomes. It is surprising that the aberrant types should outnumber the regular progeny plants without any distinct morphological difference from the latter. Similarly pentaploids of *Gossypium* were made by backcrossing the synthetic hexaploid *G. barbadense-arboreum* to *G. barbadense*, and hexaploid *G. hirsutum-raimondii* to *G. hirsutum*. Chromosomes of each plant were counted from three or more root-tip cells. Unlike the situation in tobacco, all but one of the 52 plants tested had a modal chromosome number of 65, the expected pentaploid number. Some within-plant variation was found, with the somatic number ranging from 28 to 75 or 76. The exceptional individual of 5x [(*G. hirsutum-raimondii*) × *hirsutum*] had a modal number of 70 (7 cells) with other safe counts of 61, 68, 69, 70, and 73 chromosomes. The frequent occurrence of chromosomally abnormal plants in *Nicotiana* and the remarkably uniform modal number in *Gossypium* form a sharp contrast in chromosomal stability. It is interesting to note that both crops are allotetraploids, having been cultivated for a considerable period of time, yet the chromosomes behave quite differently.

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SOME CULTURAL ASPECTS OF TWO SPECIES OF THE GENUS *Archangium* (MYXOBACTERALES). *James C. McDonald and John C. Peterson.* Pure cultures of *Archangium gephyra* and *Archangium primigenium* were achieved using several methods. The most successful method incorporated Penicillin G. (approximately 1 unit per milliliter of media) into media supporting *A. primigenium* growth and Penicillin G, Bacitracin, and Gramicidin (varying concentrations) into *A. gephyra* media. Studies undertaken to determine various physiological optima produced results indicating 30–32°C. and a near-neutral *p*H as two optima for numerous myxobacterial activities such as vegetative cell growth and fruiting body formation.

Attempts have been made to prepare a chemically defined nutrient medium for each organism with only partial success. Some 15 carbohydrates were studied as possible carbon sources in myxobacterial nutrition. Other additives, including a complete vitamin solution, nucleic acids, and casein hydrolysate, were incorporated into nutrient media in a variety of concentrations and combinations with some success. Presently active, pure cultures of both organisms are maintained on a mixed carbohydrate source plus 0.25 per cent casein hydrolysate. Vegetative cell growth is normal,

but typical fruiting bodies do not form upon these media.

THE RELATIONSHIP OF VASCULAR TISSUES IN THE MAJOR AND MINOR VEINS OF FOLIAGE LEAVES. *Brenda F. Slade.* Although a midrib of a pinnately veined dicotyledonous leaf is comparatively small in diameter at the distal end, each main lateral vein contributes substantially to its girth so that at its base it is composed of vascular tissues from all the lateral veins. This composite structure is markedly demonstrated in a developmental study. The first vascular tissue to differentiate in a primordium is the centermost strand of the midrib, which ultimately extends to the tip. Shortly afterward at the base of the primordium similar strands of the main lateral veins differentiate parallel with the first strand, but each in turn diverges from its parallel course to become the first strand of a main lateral vein in the lamina. Each of these strands is the "center" in relation to subsequent differentiation of vascular tissues in the vein. These tissues develop in the following manner:

1. Strands are added parallel with the original one so that the vein increases laterally in girth. The number of strands thus formed can be correlated with both the amount of intercalary growth and the number of minor veins produced in the intercostal panels.

2. Initially a strand is composed of only a single file of xylem and a small cluster of phloem, but a cambial-like subdivision of the procambium between these two tissues soon forms a row of protoxylem elements and additional phloem. This vertical extension of the vascular tissues is greatest in the first-formed strand of each bundle and decreases gradually in those lateral to it. The result is the characteristic wedge-shaped mass of the primary xylem of a typical dicotyledonous vascular bundle, each consisting of rows of protoxylem elements that are frequently separated one from the other by narrow files of parenchyma.

Whereas the centermost strands in each main lateral vein can be traced right down the midrib and into the petiole (and presumably continuous with the vascular bundles of the stem), those of the higher series of veinlets are progressively shorter.

BLADE TISSUE STUDIES OF SONORAN DESERT SHRUBS. *Jane Philpott.* Anatomical characteristics of foliage leaves of some desert shrubs are compared with foliage leaves of woody plants of less arid environments. The Sonoran Desert shrubs under study are from the lowlands of California near the northern end of the Saltan Sea, where daytime temperatures are very high (often well above 100°F.) and night temperatures drop about 30°. Precipitation is erratic and meager, usually less than 5 inches annually and never in the form of snow. The southern desert is considered barren but rich in the number and variety of species. Both habitats and close-up views of the foliage leaves are shown in kodachrome. Shrub species given special attention are *Ereogonum inflatum, Frourqueria farinosa, Atriplex lentiformis, Dalea emoryii, Encelia farinosa, Petalonyx thurberi, Tarrea dicaricata, Franseria dumosa, Tidestroma oblongifolia, Dalea schottii,* and *Peuciphylleum schottii.* A few deep-rooted "trees" under study from the washes in desert canyons are *Ceridium floridum, Prosopis pubescens, Chilopsis linearis, Olnyea tesota,* and *Dalea spinosa.*

In general the scurfy leaves are very small in blade area and relatively thick (average 340 μ). They are isolateral in internal organization. Small epidermal cells with various trichomes (hairs and glands) enclose the mesophyll, which is predominantly palisade. The veins are closely spaced (103 μ). In some species most of the chloroplasts are concentrated in well-developed bundle sheaths. Only a few of the leaves have bundle sheath extensions. The stomata are well spaced but nearly equal in number on the upper and lower surface.

ASPECTS OF NEGATIVE CRYSTAL GROWTH IN QUARTZ CRYSTALS FROM THE EASTERN PIEDMONT AREA OF NORTH CAROLINA. *William J. Furbish.* A study was conducted on inclusions of some crystal quartz. The quartz was formed in fracture zones of the county rock through deposition by solution.

Many of the negative crystal structures of the host quartz crystals contained clay or other detrital material. The deposition of this detrital material on a surface area of the host crystal gave rise to growth stoppage and initiated negative crystal structure formation on that surface. Continued growth of the host crystal took place around and over the blanket of detrital material, completing the negative crystal structure.

Negative crystals which apparently contain only fluid or fluid-gas were also present. Their origin is explained as being similar to that of the ones containing detrital material. Growth stoppage on an area of the host-crystal growth surface was brought about by absorption of

ions or complexes present in the quartz-crystal-forming solution. Subsequent crystal growth around and closure over the area completed the negative structure.

Both types are a result of host-crystal-growth interruption of a growth model adopted from Bunn and Emmett.

All primary inclusions in quartz formed by solution, with few possible exceptions such as those formed along lineages, have their origin as negative crystal structures by the process described. The included material may only initiate the formation of a negative crystal structure or modify the shape after its initiation.

STRUCTURAL GEOLOGY OF THE HANGING ROCK AREA, STOKES COUNTY, NORTH CAROLINA. *Barry A. Centini.* A geologic investigation is being made of the Hanging Rock State Park area in Stokes County, North Carolina. Quartzites are the chief rock types in the park area. Gneisses and schists completely surround the quartzites. No sedimentary or igneous rocks have been encountered in the mapping.

Tentative results based on discontinuity of lithology and structure seem to indicate a low-angle-thrust fault. The gneisses and schists appear to have been thrust over the quartzite, which now appears in a fenster.

No definite correlation of rock units and structure of the Hanging Rock and surrounding quartzite areas with the regional geology has been made. However, structurally and lithologically this area appears similar to the southeastern corner of the Grandfather Mountain Window. The quartzites and gneisses and schists of the Hanging Rock area resemble the Chilhowee Group and Inner Piedmont rocks respectively of the Grandfather Mountain area. Furthermore, the thrusting of gneisses and schists over quartzite is similar in the two areas.

ROLE OF CLAY MINERALOGICAL ANALYSES IN SOIL GENESIS INVESTIGATIONS. *L. A. Nelson and R. J. McCracken.* Soil scientists have long been interested in the clay fraction of soil materials from a plant-nutrition standpoint. Clay mineralogical studies have also been applied in soil-genesis investigations in recent years.

There are a number of methods currently used in clay mineralogical investigations for studying soil genesis. Several different types of determinations are often made because they are complementary and provide information which would not be available from any one determination alone.

X-ray diffraction analysis is perhaps the most commonly used method for studying the crystalline components of the clay fraction. Inferences as to types of minerals present, as well as estimates of quantities present, can be made from X-ray data.

Differential thermal analyses are useful in studying thermal reactions of the clay fraction.

Clay mineral identification is possible by this method because specific minerals have characteristic thermal properties (water losses, phase changes, etc.)

Cation exchange capacities are useful in detecting materials having high negative charge (montmorillonites, vermiculites, amorphous materials). Clay mineral identification by this method is more difficult for clays having lower charges. Differential dissolution analysis is useful in isolating mineral components for more detailed studies by other methods. Electron microscopy is useful in characterizing the size and shape of clay particles; therefore it can be used in clay mineral identification.

Several important soil genetic inferences can be drawn from clay mineralogical data. These are based upon the assumption that past weathering patterns are a result of a number of factors known to affect clay mineral suites present in soil materials. Some of these factors are depth relative to surface, particle size, and natural drainage.

For example, in many North Carolina soils, a maximum of a clay mineral (or clay mineral complex) with a basal spacing of approximately 14.3 Angstroms is noted in the upper horizons. The concentration of this material, possibly to an intergradient vermiculite-chlorite with partial interlayering of aluminum, decreases with increasing depth in the soil profile. In contrast, the proportion of kaolinite increases, as inferred from peak heights or X-ray diffractometer tracings. Also, this 14A mineral is concentrated in the coarse clay fractions. Studies designed to supply information on these occurrences are needed.

With respect to natural drainage, we have noted that montmorillonite and other three-layer clays tend to occur in poorly drained soils. However, soils with good natural drainage formed from similar parent material possess a kaolinite-14A mineral suite.

GEOLOGY OF THE PYROPHYLLITE DEPOSITS AT GLENDON, MOORE COUNTY, NORTH CAROLINA. *James F. Conley.* A study of the pyrophyllite deposits located approximately one and one-half miles north of Glendon, Moore County, North Carolina, was made as part of an areal

mapping program of Moore County. From this study it was learned that the deposits occur in the shear zone of a N 60° E trending reverse fault which dips to the northwest at 50°. The country rock surrounding the deposits is an argillaceous slate of the Carolina Slate Belt Sequence. The pyrophyllite deposits are zoned. These zones are classified as follows: outer magnesium and iron-enriched zone, potassium or alkali zone, and an inner high-alumina zone. The origin of the deposits is somewhat in doubt. In the author's opinion the only mechanism which could account for segregation of different suites of minerals into restricted zones in the confined limits of the shear zone of a fault would be hydrothermal replacement.

WHAT IS THE FUTURE OF THE GEOLOGY SECTION IN THE NORTH CAROLINA ACADEMY OF SCIENCE? *Stephen G. Conrad.* Since the Geology Section of the North Carolina Academy of Science was first organized in 1941, it has been subjected to varying degrees of support from geologic profession in the state. For instance, in some years the annual program has been crowded by as many as twelve papers, while at other times as few as three papers have been presented.

At the present time, there are more geoscientists engaged in a wider field of geologic research in North Carolina than at any other time in the state's history. In view of the present level of geologic endeavor, which promises to become even broader in scope in future years, it appears that the Geology Section is not serving the geoscience profession in the state to its full potential. However, with proper support and enthusiasm the Geology Section could become a very definite asset to the geoscience profession in the state.

Two ways in which the section perhaps could serve best are by (1) encouraging graduate students to present the results of their investigations to the section, which would give the students an opportunity to appear before a relatively small but interested group; and (2) by the presentation from time to time by faculty members at the universities and other geoscientists working in the state of progress reports on their individual research projects. This would allow other geoscientists in the state an opportunity to keep abreast of contemporary work and an opportunity for the workers to get the reaction and constructive criticism of their fellow geoscientists.

TEXTBOOKS IN USE IN NORTH CAROLINA COLLEGES AND UNIVERSITIES. *Marshall Helms.* A listing of the texts used in undergraduate physics courses of the colleges and universities in North Carolina was compiled and presented in tabular form. Information was obtained from twenty-eight institutions and includes all but one which have sizeable physics programs, such as a minor or major in that department. Besides the one missing, replies from two other colleges were too late to be included in the paper as presented to the Physics Section. The tabular display offers ready information about the books which are used as texts in the general courses as well as in the more specialized courses, which include (1) intermediate and advanced mechanics; (2) heat and thermodynamics, (3) electricity and magnetism; (4) light and optics; (5) modern physics, including atomic and nuclear physics; (6) electronics; and (7) miscellaneous subjects such as astronomy, laboratory arts, and meteorology. Many text are used: in general physics alone (excluding brief courses as Household Physics, etc.) nineteen different texts were used in the twenty-eight colleges involved in the tabulated results. However, for optics only three texts were used. Books used in heat were copyrighted in 1948, 1950, 1953, and 1960. These dates suggest that physicists in their writing have neglected the area of heat while overproducing in general physics and other areas, such as modern physics. Published laboratory manuals are little used; for general physics 42 per cent of the courses have such manuals while 58 per cent use laboratory directions and sheets prepared by the local staff. In the advanced courses published manuals are used for only 9 per cent of the laboratory work, while for 91 per cent of such work the local staff produces the lab sheets or selected references.

The tables also give some indication as to the undergraduate physics programs of the several colleges by presenting their courses and credits, the number of hours required for physics major or minor, and the number of physics majors graduated in the two years 1957–59.

The paper concludes with a bibliography of eighty-two texts used in physics classes in North Carolina colleges.

IN-SERVICE INSTITUTE AS AN AID TO THE HIGH SCHOOL PHYSICS TEACHER. *Sherwood Githens, Jr.* The teachers in an in-service institute entitled "Laboratory Physics for Teachers," which was given on Monday nights, were asked to write not a commentary on the course, but rather to itemize specific instances or ways in which participation in this course had been

of direct benefit in their teaching. In the material received, eighteen different aspects of aid were found and listed. This list, read during the talk, includes a number of benefits not anticipated when the course was conceived.

A REPORT ON THE RECENT MATHEMATICS-PHYSICS ACADEMIC YEAR INSTITUTE CURRICULUM CONFERENCE AT WASHINGTON UNIVERSITY. *W. E. Haisley.* The central concern of this conference was the interaction between high school courses in the "new mathematics" (SMSG, Ball State, Illinois) and the "new physics" (PSSC), and the relation of each to the academic-year institute-training programs for high school teachers. In general the new high school materials are more nearly completely assimilated by the institute curricula in mathematics than by those in physics. Regarding the effect of the new mathematics courses on physics teaching, there seemed general acceptance among delegates in both disciplines that in the future an increasing proportion of necessary mathematics will have to be taught within physics curricula.

A NEW METHOD FOR DETERMINING THE TRUE COUNT RATE FROM A NUCLEAR DETECTOR. *Wesley O. Doggett.* A new technique for determining the true count rate from a radiation detector is proposed. The method is applicable to measuring the count rate from a constant source of radioactivity and is essentially free from dead time losses associated with the detector. Furthermore, the output pulses to be registered are regularized, and the detector need not be sensitive during the entire counting period. The method, therefore, has the potential for increasing the count rate capability of a scaler and for extending the useful life of a Geiger tube. The experimental technique consists of dividing the time of a measurement into a large number of intervals, each of equal length and somewhat longer than the resolving time of the detector. The probability P_o that no counts are detected in an interval is determined by dividing the number of such intervals by the total number of intervals. According to the Poisson distribution, the average number of counts per interval is $-\ln P_o$, from which the average count rate can be determined. Experimental results obtained by Mr. Bou-Loong Ho with a calibrated source showed that the count rate determined by this zero probability method agreed with the true calibrated count rate (experimental accuracy 0.5 per cent) even when the dead time loss rate was as high as 15 per cent.

RADIATION REACTION IN A GRAVITATIONAL FIELD. *Robert W. Brehme.* Contrary to what might be expected from both the classical theory of radiation from an accelerated charge and the equivalence principle of general relativity, a charge which is permitted to move freely under the action of a gravitational field indirectly experiences radiation damping. However, the charge does not follow the geodesic, not solely because it radiates energy in the usual sense, but because the gravitational field is able to scatter the electromagnetic field back onto the particle, causing the particle to deviate from its geodesic. The deviation causes non-vanishing terms to appear in the expression for radiation damping, implying that radiation is emitted by the charge.

COSMOLOGICAL SPACES ADMITTING GEODESIC CORRESPONDENCE. *B. E. Leonard and W. R. Davis.* The dynamical aspects of the possibility of two or more distinct Riemannian spaces admitting the same paths of motion (geodesic correspondence) has been previously investigated by others. In view of the fact that models of cosmology and matter singularities in the general theory of relativity are of a spherically symmetric or an isotropic nature, spaces having the following type fundamental form have been studied to determine what restrictions the demand of geodesic correspondence places on the g_{ij}'s.

$$ds^2 = g_{ij}(r,t)\,[dr^2 + r^2\,d\vartheta^2 + r^2\sin^2\vartheta\,d\varphi^2] + e^2\,dt^2$$

Necessary and sufficient tensor equations which are equivalent to those first formulated by Weyl were utilized. It was found that the most general spaces admitting geodesic correspondence are of the form

$$ds^2 = \frac{-e^{g(t)}}{(1 + r^2/4R^2)^2}\,[dr^2 + r^2\,d\vartheta^2 + r^2\sin^2\vartheta d\varphi^2] + c^2dt^2$$

where e^g must satisfy the differential equation

$$\frac{d^2g}{dt^2} \pm 2k^2\,e^{-g} = 0,$$

Solutions of this equation are of four forms

$$\begin{aligned} e^g &= e^2bt \text{ if } k = 0 \\ &= (kt + a)^2 \\ &= \sin^2(kt + a) \text{ or } \cos^2(kt + a) \\ &= \sinh^2(kt + a) \text{ or } \cosh^2(kt + a) \end{aligned}$$

This space is the same as one obtains in the construction of the Friedman-Lamaitre non-statie cosmological models as discussed by Tolman, and specialized by the differential

equation to be the condition as shown by Takeno that the space be static and of constant curvature. It follows that any nonstatic space admitting geodesic correspondence will necessarily be more general than the Friedman-Lamaitre cosmological models. Also we have found that the Schwartzchild solution does not admit geodesic correspondence. More general nonstatic spaces are to be investigated and presented in subsequent publications. If spaces of physical interest admit geodesic correspondence one would in principle need for example a test particle with spin to detect the difference between such spaces by observing the test bodies' motion. This would follow from the work of Bergmann and Schiller, and also of Papapetrou, which shows that the spin angular momentum tensor of such a test particle would couple with the components of the curvature tensor in the equations of motion. The physical implications of this are also to be investigated.

A GENERALIZED VARIATIONAL METHOD FOR SOLVING DIFFERENTIAL EQUATIONS OF PHYSICS. *Alfred L. Mowery, Jr., and Raymond L. Murray.* This paper is devoted to the exposition and illustration of a technique the authors have designated as the generalized variational method (GVM). The analysis is based on the variational approach and is an outgrowth of investigations in the hypercircle method. In essence, the GVM consists of considering the trial functions that appear symmetrically (quadratically) in a positive-definite variational principle as independent functions. A proposition was proved to demonstrate generally that the approximate eigenvalue obtained from the GVM is at least as accurate as the geometric average of the associated approximate eigenvalues. Also, a conjecture was proposed that the accuracy of the generalized variational eigenvalue is comparable to that of a variational result employing a trial function incorporating the dimensionality of both associated trial functions. The application of the GVM to the perturbation-variational method yielded results that firmly establish the method. The generalized method completes the perturbation-variational method by providing the formerly missing even-order approximate results. For illustration, the GVM was employed to solve a bare reactor with a grey control sheet. Using Rayleigh-Ritz optimized cosine series and optimized pyramid functions as associated solutions, the generalized variational eigenvalue accuracy indicated the effective combination of the dimensionalities of the associated trial functions.

ANALYTICAL STUDIES OF NEUTRON SLOWING AND DIFFUSION. *Donald E. Selis and Raymond L. Murray.* A detailed analysis of a previous age-theory derivation for a bare, finite, cylindrical nuclear reactor with a fully inserted central control rod is presented. The work done by F. H. Murray, referred to frequently in the literature, is greatly amplified and includes the mathematical steps leading to his results. In order to determine the validity of the physical and mathematical assumptions of this paper a slab reactor with a central control zone was investigated using both age-theory and two-region techniques. The results of the age derivation are used in calculating the fast-extrapolation distance, d_1, into the control zone. A critical reactor is determined by two-group methods and the resulting fuel loading and other constants are used to determine the age-theory fast-extrapolation distance. The difference between the age and two-group values is greater than expected, which is associated with the strict conditions placed on the age-theory derivation. In the course of the analysis special functions that are generalizations of the error function were studied, along with the properties of integrals over Bessel functions of various kinds and orders.

A LABORATORY EXPERIMENT USING NEUTRON THRESHOLD DETECTORS. *D. H. Martin.* A modification of the method of Jablowski and Di-Meglio for measurement of fast-to-thermal neutron flux is described. A measurement using this method is utilized as one of a series of reactor experiments in a nuclear reactor laboratory course at North Carolina State College. The procedure involves irradiating an aluminum foil in a neutron flux, then counting the resulting gammas in a single-channel spectrometer, observing first the 2.32 min. half-life associated with thermal activation (1.8 Mev) and then the 9.5 min. half-life from fast (above the 2.1 Mev n, p threshold) activation (0.84 Mev). A calibration factor for comparing the two activities is obtained from irradiated manganese, which produces gammas almost identical to the two of interest and of a known ratio.

Typical values of the fast-to-thermal flux in the center of the North Carolina State water boiler reactor (NCSCR IV) are estimated from student data to be about 1.5. Further improvement of this method as well as its use in measurement of reactor spectra is planned as part of master's thesis research in the immediate future.

A NEW STRIGEOID TREMATODE (*Diplostomatidae*: *Alariinae*) FROM FLORIDA RACCOONS. *Reinard Harkema and Grover C. Miller.* Examination of 19 raccoons from Glades County, Florida, in September, 1960, revealed the presence of an unknown strigeoid trematode. Five of the 19 raccoons had light to moderate infections of this parasite in the small intestine. These specimens are sufficiently different from any known form to constitute a new genus. They are characterized by an elongated bilobed holdfast organ which extends to or beyond the oral sucker. There is similarity to the genus *Pharyngostomoides* Harkema, 1942, in the parallel position of the testes and in general body form. The major features which separate this strigeoid trematode are the position and structure of the holdfast organ, the forward location of the testes, the large recess in the genital atrium, and the much larger size.

(Supported by Grant E-3209, National Institutes of Health.)

PRELIMINARY STUDIES ON THE LIFE HISTORY OF *Heterobilharzia americana* PRICE, 1929. *Nathan D. Greene. Heterobilharzia americana* Price, 1929 (Trematoda: Schistosomatidae) has been reported from *Lynx rufus floridanus* in Florida (Price, 1929, Proc. U. S. Nat. Museum, **75**: 1–39), *Procyon lotor* in Texas (Price, 1943, Proc. Helm. Soc. Wash. **10**: 85–87), and from *P. lotor* in eastern North Carolina (Miller and Harkema, 1960, J. Parasitol. **46**(2): 206). The life history of this blood fluke is apparently similar to that of other members of the family Schistosomatidae. Eggs are voided in the feces of the infected animal and hatch within minutes after deposition in water. The miracidium penetrates a snail of the genus *Lymnea* (*Pseudosuccinea*) in which development requires approximately four weeks. Emerging cercariae gain entry into the final host by means of direct penetration. Studies of the morphology of the cercaria and developmental stages in the molluscan intermediate host are in progress.

(Supported by Grant E-3209, National Institutes of Health.)

MICROSTRATIFICATION OF THE TOMOCERINAE (COLLEMBOLA) IN A BEECH-MAPLE FOREST OF EASTERN NORTH CAROLINA. *Clifford B. Knight.* The soil profile of a beech-maple stand was randomly sampled biweekly and the Tomocerinae separated from four recognizable strata. *Tomocerus flavescens separatus* and *Tomocerus lamelliferus* were found most frequently in the "F" stratum (A_{oo}) and "H" stratum (humus, A_o). Most specimens measured from 0.75 mm. to 1.5 mm. in length, smaller specimens were more numerous during the hiemal period for both trivial groups. Largest populations for both species occurred in the late vernal and early estival periods, while the smallest populations for *T. flavescens separatus* were during the late estival and early hiemal periods. *Tomocerus lamelliferus* exhibited only one major population decline during September and early October collections. Possible food scarcity and thermal extremes were believed to be the most important factors inhibiting population growth.

PATTERN OF EMERGENCE IN THE DRAGONFLY *Tetragoneuria cynosura*. *Paul E. Lutz.* Since rates of emergence in dragonflies have been investigated adequately in only a few forms, a study has been made of the pattern of emergence in *Tetragoneuria cynosura* in the spring of 1960 and 1961 at University Lake near Chapel Hill, N. C. A shore line measuring 225 feet and representing a variety of habitats was examined with approximately the same degree of effort for exuviae of this species in both years.

In 1960 the first east nymphal skin was collected on April 14, initiating an emergence period of 23 days. The rate of transformation increased rapidly and reached an explosive peak on April 23. Of the 1,711 exuviae collected in 1960, approximately half were obtained during the first week and more than 85 per cent by the ninth day of the emergence period. In 1961 the emergence period began on April 5, nine days earlier than in the preceding year. Based on only 524 exuviae, the emergence rate remained quite low until a small peak was recorded approximately three weeks after the beginning of the emergence period. There is evidence that the earlier and more prolonged emergence period and the temporally displaced emergence peak in 1961 may be attributable to climatological factors. The striking difference in numbers of exuviae collected in the two years strongly suggests a significant annual fluctuation in population size.

The diurnal pattern of emergence was investigated on April 22 during the 1960 explosive peak. Of the 384 exuviae collected at three-hour intervals on this day, 50 per cent were collected by 6:00 a.m. and 75 per cent prior to 9:00 a. m. Transformations occurred throughout the day and as late as 7:00 p. m. No diurnal differences in rate of emergence for the two sexes were observed.

From this study it was found that sex ratios in both years departed significantly from 1:1.

In 1960 the ratio was found to be 4 males: 5 females compared to a ratio of 2 males: 3 females in 1961. In comparing the sex ratio for the two years, a significant difference was noted. The data also indicate that males emerged earlier than females in both years but female exuviae were more abundant later in the emergence period. It is felt that this has ecological significance in that it provides a mechanism whereby males will be present when females return to the water to mate and oviposit.

TYPES OF INDUCED MODIFICATIONS IN THE PATTERN OF OVIPOSITION OF THE WASP. (*Habobracon*). *Daniel S. Grosch.* In addition to the standard pattern, four distinct types of egg-production curves have been obtained in experiments with an inbred braconid stock: (1) compensatory deposit late in life making up for a deficit which follows a feeding of an antimitotic agent, (2) a constant deficit throughout life after single-meal ingestion of sulfhydryl reactions, possibly indicating general debility, (3) a "dromedary" curve whose depression reflects the vulnerability of cells differentiating from oögonia to oöcyte-trophocyte units, (4) a lag phase of up to 5 days corresponding to degeneration of oöcytes. The third type is characteristic of moderate radiation exposures. Antimetabolites have given both third and fourth types with no particular pattern emerging except that the more potent analogues tend toward the fourth type of result. Among these are DON, Aureomycin, and the halogenated deoxyuridines. Thus the result may be obtained with agents influencing both amino acid and nucleic acid synthesis. A plea is made for cytological investigation, especially since there has been a shift of attention in economic entomology toward the chemisterilants. The ovarioles of different species exhibit different degrees of differentiation at the time of treatment.

REGENERATION OF THE INTESTINE OF *Ambystoma* LARVAE. *Marilyn S. Kerr* (introduced by Edward C. Horn). In an effort to determine the regenerative capacity of the larval salamander gut, intestines of *Ambystoma opacum* larvae were transected approximately four millimeters posterior to the pylorus. No attempt was made to orient the gut ends, which were returned to the body cavity immediately after transection. Histological studies of animals killed at specific intervals, as well as gross microscopic observations of living and fixed animals, were made on experimental and control larvae. The sequence of events leading to regeneration was found to be (1) initial widespread extension of mucosal and serosal epithelial tissues; (2) establishment of adhesions between the free ends of these tissues with coelomic organs or tissues other than gut; (3) fusion of the mucosal layers from the two ends of the gut; (4) blastema formation, growth, and differentiation into gut-wall tissues. Blastema development depended upon adhesion of a loose end of the gut, not only with the other gut segment, but also with some other tissue, the dissociation of which contributed large morphologically undifferentiated cells to the blastema-like mass accumulating at the region of attachment. The blastema was formed by cells of the dissociated gut serosa, submucosa, and muscularis; dedifferentiated striated muscle cells, as well as peritoneal and connective tissue cells, also arising from adhered visceral organs, were incorporated into blastemata developing at sites of body-wall attachments. Reformation of the gut lumen occurred by continued fusion of the nondissociating mucosal layers of the two gut segments. Blastemal cells gave rise to submucosal and serosal tissues, the latter also forming by migration of rapidly proliferating peritoneal cells of the adjacent intact gut regions. Muscularis, the last tissue to be formed by alignment and differentiation of blastemal cells, was incomplete to some extent in all larvae studied histologically. Ileostomies, located at sites of adhesions between gut end and body wall, occurred in 33 per cent of the experimental larvae.

GERMINAL LOCALIZATION AND MORPHOGENESIS OF MIDGUT DERIVATIVES IN THE SALAMANDER *Ambystoma punctatum*. *Thomas M. Harris.* Preserved eggs at representative developmental periods from oöcyte to larval stages were cut in half along the median sagittal plane to form hemisections. Such hemisections of unfertilized eggs show an internal organization of the cytoplasm into three distinct areas, each with its characteristic pigmentation. After fertilization the internal pigmentation pattern changes markedly in what is believed to be an example of oöplasmic segregation.

As a result of the oöplasmic segregation, a cap of densely pigmented vegetal plasm that has not previously been reported becomes localized immediately over the vegetal pole of the egg. This vegetal plasm is carried up through the center of the yolk mass by cortical ingression associated with the early cleavage furrows. By the late blastula stage the pigmented vegetal plasm is distributed as a cone through

the yolk mass. The base of the cone occupies the vegetal pole of the egg while the apex lies immediately beneath the center of the blastocoel floor.

Vital-staining studies of the central vegetal-pole cells indicate that during gastrulation they migrate through an arc of 180° and come to rest over the apex of the pigmented cone. They then invaginate into the center of the cone, which during gastrulation has become oriented in an anterior-posterior direction. Although some of the invaginated central endodermal cells remain attached to the surface of the archenteron floor, others penetrate the full depth of the cone. These cells and their progeny thus form a path through the center of the yolk mass. They have been observed to be instrumental in the formation of the intestinal lumen in later development.

The above observations confirm the assumption of Balinsky that the lumen of the salamander gut is not derived from the primitive midgut, but is rather a new cavity which arises through the center of the yolk mass. It is further indicated that the cells which are instrumental in the lumen formation are the direct progeny of cells originally derived from the localized vegetal plasm of the uncleaved egg.

(This investigation was carried out during the tenure of a predoctoral fellowship from the Division of General Medical Sciences, United States Public Health Service.)

THE EFFECTS OF REMOVAL OF PORTIONS OF KIDNEYS ON CELLULAR PROLIFERATION IN TUBULAR EPITHELIA OF REMAINING KIDNEY TISSUE. *Charles E. McCreight.* It has been demonstrated that surgical removal of one kidney is followed by significant increase in mitotic activity in the tubular epithelia of the remaining kidney. This study was designed to determine whether the amount of kidney epithelium removed could be correlated with the hyperplasia occurring in the remaining tissue.

One-half of each kidney of 4- to 7-month-old male rats in one group was removed by electrocautery. One complete kidney and one-half of the other was removed from the animals in another group. All animals were sacrificed 50 hours postoperatively.

Mitotic figures in proximal convoluted tubules of control kidneys were infrequent constituting 0.10 per cent of the cells. In the remaining kidney of young rats 72 hours following unilateral nephrectomy (McCreight and Sulkin, 1959) the mitotic count comprised 0.69 per cent of the cells of the same epithelium. In remaining half-kidneys following removal of one-half of each organ, cells of the proximal convoluted tubules undergoing division comprised 0.66 per cent. Mitotic activity, therefore, was found to increase to about the same extent in two remaining half-kidneys as in one remaining entire kidney.

In remaining single half-kidneys, where the kidney tissue had been reduced to approximately one-fourth its original amount, mitotic activity in proximal convoluted tubules increased tremendously. Here the cells in mitosis comprised 1.84 per cent. Mitotic figures were nearly three times as frequent as in the same tissues following removal of one-half the kidney complement. Mitotic activity in the tubular epithelia of remaining kidney tissue does not increase in direct proportion to the amount of kidney tissue removed. The rate of mitotic increase appears to be enhanced upon removal of greater amounts of tissue.

MAINTENANCE OF THE UNHYDRATED STATE OF EGG JELLY IN THE *Rana pipiens* REPRODUCTIVE TRACT. *Thomas W. Lee* (introduced by Edward C. Horn). The jelly coats of oviducal and uterine amphibian eggs do not normally imbibe water from the body fluids, although rapid swelling of the jelly occurs when contact with water is made either within or without the reproductive tract. Therefore, it is reasonable to suppose that within the reproductive tract there is an inhibitory or isolating mechanism preventing the uptake of water by the jelly coats. The studies carried out indicate that none of the following factors are involved in the prevention of jelly swelling in the reproductive tract: *p*H, salt concentration, or water removal from the lumen. Permeability studies utilizing hypertonic sucrose solutions (10 per cent) containing known amounts of Evans Blue, the change in concentration of which may be determined colorimetrically, have shown the ovisac to be permeable from outward to inward to Amphibian Ringer's solution. However, when an ovisac is filled with eggs alone, there is no osmotic passage of body fluids, or Amphibian Ringer's, into the ovisac. This indicates that the eggs are osmotically isolated when they are contained within the ovisac. Coelomic injections of Evans Blue into nonovulating pituitary-activated females, as well as coelomic injections of Amphibian Ringer's into ovulating females, have shown that no coelomic fluid gains entrance to the oviduct. Therefore, it appears that osmotic isolation of uterine eggs, coupled with some unknown barrier to the entry of body fluids into the ostia of the oviducts, is operative in

maintaining the unhydrated state of egg jelly in the *Rana pipiens* reproductive tract.

DIFFERENTIATION OF HYDRA INTERSTITIAL CELLS IN CULTURE. *Maximo Cerame-Vivas and C. G. Bookhout.* Interstitial cells of *Pelmatohydra oligactis* (Pallas) were isolated by first separating the epidermis from the gastrodermis. This was done by placing hydras in a 0.5 per cent solution of di-sodium ethylene-diaminetetraacetate in Loomis culture water, which dissociates the mesolamella. The hydras ruptured at the pedal disc and the gastrodermis was extruded. The epidermis contracted and remained curled below the hypostome. Separation was then completed by using glass needles and small wire loops. Cells of the isolated epidermal layer were carefully separated in a "Teflon" tissue homogenizer and the cell suspension was submitted to centrifugation in an ordinary hand centrifuge. The interstitial cells were isolated from the suspension and hanging drop preparations were made. The culture media provided were extracts of the gastrodermis and from the epidermis. These were obtained by freezing and thawing separated cell layers. Differentiation of the I-cells usually took place within from twenty-four to thirty-six hours, but, regardless of the medium provided, differentiation took place only toward the formation of cnidoblasts and development of nematocysts. No differentiation into other gastrodermal or epidermal cell types was observed.

SIAMESE PIGEONS. *Robert W. Prichard and Torguan Tan.* The urban and domesticated pigeons of Thailand (Siam) have both similarities to, and differences from, the pigeons found in similar situations in the United States. Taxonomically they are all *Columba livia domestica*, but the varieties under this heading are fewer than the approximately 70 listed by Levi (*The Pigeon* [The Levi Publishing Company: Sumter, S. C., 1957], pp. 37–238).

From the standpoint of novelty the Laugher is perhaps the most interesting Siamese pigeon. This variety, unknown in the United States but intermittently imported into England only to die out, derives its name from its voice, which is quite unlike that of other varieties. These birds were apparently brought to Thailand from Mecca by Thai Muslims making their pilgrimage to that religious center. The importation continues, for the Muslims believe that certain Laugher voices are "lucky," and they will pay large sums of money at auction for the privilege of eating a live Laugher with a "lucky" voice, which act is said to transfer good fortune to the eater. The vast majority of these birds are self white, clean headed, and clean legged. Only three black Laughers are known to us. The genetics of this type of voice are presently under study.

Thai common pigeons, living as scavengers on buildings, consist of two principal varieties. The most numerous are the blue checked and barred pigeons similar to those in the United States. The second group consists of large birds, usually muffed and often crested, resembling the Duchess pigeon as seen in the United States. These birds are probably related most closely to the Lahore, a variety said to have been developed in the Indian city of that name.

Fantails, long a popular variety in Europe and the United States, appear in Siam both crested and muffed, characteristics which have not been present in the West for sixty years or more. Most Siamese fantails are self white, and rarely do they appear in the various colors and patterns popular elsewhere. As in the case of the Lahore-type common pigeon, there is a similarity to the Indian variety of contemporary fantail. Most authorities consider India the probable original home of this very popular pigeon.

Chinese Owls, similar to that variety as it is seen in the West, are common, both in the hands of fanciers and flying at large. At one time they were used for short races, but the introduction of British racing homers spelled the end of that practice.

Rice, corn, and mung beans are the usual grains fed pigeons in Siam. Both polished and unhusked rice are used, with apparently satisfactory growth and development. Little attention is paid to mineral mixtures, and squab production seems to improve when an adequate preparation is fed. Rooftop lofts, as in large European and American cities, are common, with teak a favorite building material.

(Aided by Grant H-4352, United States Public Health Service.)

SOME FISHES FROM THE AREA OF CHINCOTEAGUE BAY, VIRGINIA. *L. M. Outten.* Collections of fishes were obtained during September, 1949; June and September, 1950; and September, 1960, near the shore and in the first stream south of Red Hills on Chincoteague Bay, Virginia. The following species were among those present in the area: *Anguilla rostrata* (LeSueur), *Cyprinodon variegatus* Lacépède, *Fundulus diaphanus* (LeSueur), *F. heteroclitus* (Linnaeus), *F. majalis* (Walbaum), *Eucinostomus*

pseudogula Poey, *Orthopristis chrysopterus* (Linnaeus), *Bairdiella chrysura* (Lacépède), *Gobiosoma ginsburgi* Hildebrand and Schroeder, *Mugil curema* Valenciennes, and *Menidia menidia* (Linnaeus).

OBSERVATIONS ON THE SPAWNING COLORATION AND BEHAVIOR OF SOME CYPRINID FISHES. *L. M. Outten.* Fishes of the family Cyprinidae are generally well represented in tributary streams of the upper Tennessee River system. About the time of spawning, males may have more pronounced coloration and engage in characteristic behavior.

Notropis rubricroceus (Cope) males at spawning time have the snout and lower jaw bright red and much of the head, dorsal and lateral parts of the body reddish. A heart-shaped area on the top of the head, the mid-dorsal line, and the lateral longitudinal bands are greenish yellow. The fins are golden yellow. Spawning occurs over nests of coarse gravel and small stones collected and arranged by males of *Hybopsis micropogon* (Cope). Spawning groups were observed at times from April 29 through July 21.

At spawning time, males of *Notropis coccogenis* (Cope) show pronounced reddish orange on the tip of the snout and along the margin of the upper jaw, in a triangular area at the anterior base of the dorsal fin, and in a small spot at the base of each pectoral fin. Spawning was observed over nests of *H. micropogon.*

Males of *Notropis leuciodus* (Cope) have at spawning time reddish coloration on the head (well developed on the snout and lower jaw), anterior part of the body, and fins (pectoral especially, also basally on the dorsal, pelvic, and caudal). While the pigmentation sometimes approaches orange-red, it may at times resemble a subdued rose or strawberry-juice tint. Territory-holding and spawning aggregations have been observed over nests of *H. micropogon.*

At the time of spawning, large mature males of *Notropis galacturus* (Cope) have the dorsal parts of the body dark blackish gray, the lateral and ventral regions silvery to milk-white with prominent characteristic white areas at the base of the caudal fin. The pectoral and dorsal fins have a reddish tinge distally, the caudal pronounced orange to crimson coloration in a wide band with a faint suggestion of white along the posterior edges, more prominent on the tips of the pectorals and caudal, the latter often with flashing white tips on the upper and lower outer margin. Spawning, usually at the lower surface or side of submerged objects, was observed at times during the period from May 25 through June 28.

AMMONIA AND UREA EXCRETION IN SOME PAEDOMORPHIC URODELES. *Richard C. Bruce.* Ammonia and urea excretion was studied in four species of paedomorphic urodeles: *Amphiuma means tridactylum* Cuvier, *Necturus maculosus maculosus* (Rafinesque), *Siren intermedia intermedia* LeConte, and *Siren lacertina* L. The technique employed involved the collection of excretory products in a known volume of water in which an animal was kept; then analyzing a sample of this water after a given period of time for ammonia and urea, using a microdiffusion method.

Necturus excretes the greater fraction of its waste metabolic nitrogen as ammonia, which gave rise to 60–90 per cent of the total ammonia-urea nitrogen in the excreta. In fasting *Necturus* the ammonia-urea ratio of the excreta increased with starvation. *Siren intermedia* excretes 80–95 per cent of its ammonia-urea nitrogen as ammonia, while for *Siren lacertina* the corresponding range was 35–55 per cent. The nitrogen partition of *Amphiuma* showed great individual variation, although ammonia usually predominated as a source of excretory nitrogen. However, *Amphiuma* which were fed large quantities of liver excreted 70–80 per cent of the total ammonia-urea nitrogen as urea during the early part of subsequent fasting. With extended fasting and the reduction of total ammonia-urea excretion, the excretory pattern reverted to the ammoniotelic condition.

The results suggest that the nitrogen partition of the excreta of these animals is quite labile. It is proposed that the high level of ammonia excretion exhibited by these four species is not necessarily a reflection of the condition of neoteny *per se,* but rather of the aquatic environment which all inhabit.

RANDOM OBSERVATIONS OF FRESH-WATER INVERTEBRATES. *Tom Daggy.* A description of the feeding of *Microstomum sp.* (Platyhelminthes: Turbellaria) on brown hydra is given, and the incorporation of the coelenterate nematocysts in the flatworm epidermis demonstrated. The nematocysts are shown to be functional in the worm.

Attention is called to three turbellarians from a vernal pool in Cabarrus County, North Carolina. These are *Mesostoma vernale* Hyman, 1955; *Dalyellia viridis caroliniensis* Hyman, 1955; and *Hymenella retenuova* Castle, 1941.

This vernal pool is the type locality for the first two.

The occurrence of *Prostoma rubrum* (Leidy), 1850 (Nemertea), and *Actinosphaerium eichhornii* var. *majus* Penard, 1902 (Protozoa, Actinopoda) is recorded for a permanent artificial pond in Mecklenburg County, North Carolina.

THE INFLUENCE OF THERMAL AND OSMOTIC STRESS ON THE CILIARY ACTIVITY OF GILLS OF LAMELLIBRANCHS. *F. John Vernberg, David E. Schneider, and Carl Schlieper.* Sedentary animals living in the intertidal zone experience wide daily and seasonal fluctuations in environmental factors undertaken to compare differences in survival of cilia of gills of lamellibranchs at high temperature and various salinities. *Crassostrea virginica* and *Modiolus demissus* are sessile species as adults, while *Aequipecten irradians* is mobile.

Thermal resistance: Gill tissue from *Crassostrea* and *Modiolus* could withstand higher temperatures than tissue from *Aequipecten*, i.e., at 38°C. gill tissue of *Aequipecten* survived 34 minutes whereas gill tissue from *Crassostrea* and *Modiolus* withstood 45° for 56 and 35 minutes respectively. Cold acclimation (10°C.) greatly reduced the survival time of tissue from the sessile species but had little effect on *Aequipecten*. This difference in thermolability might be important in adaptation to seasonal changes.

Osmotic resistance: *Aequipecten* gill tissue was unable to withstand as low salinity as *Crassostrea* and *Modiolus*. Tissue from warm-acclimated animals (25°C.) could survive at lower salinities than cold-acclimated individuals.

The possible significance of these results to the distribution of these species was discussed.

(Supported by a grant from Duke University Research Council)

LARVAL DISTRIBUTION OF THREE SPECIES OF BALANOMORPHA IN RELATION TO SOME CHEMICO-PHYSICAL FACTORS. *Murad-B. M. Mohammad.* The six hundred quantitative plankton samples on which the current investigation is based were procured usually at weekly intervals from May, 1960, through April, 1961, at a number of stations along the main channel of Newport River, Carteret County, North Carolina.

The field observations, supplemented by laboratory experiments, indicate that the upriver limit of distribution of the larvae is determined mainly by low salinities. The planktonic larval stages of *Balanus improvisus* occur in a salinity as low as 0 °/oo. The extreme low limit of salinity tolerance in larvae of *B. eburneus* is higher than for *B. improvisus* (about 8 °/oo) and even higher still (13 °/oo) for *B. amphitrite*. Larvae of all three species thrive successfully in strictly marine environments (salinities 32 to 36 °/oo).

Owing to wind-driven turbulence and tidal currents, an extensive mixing of surface and bottom water occurs in the broad part of the estuary and, as a consequence, the larval stages of the three species of barnacles are rather uniformly distributed. In the sheltered narrow segment of the river the behavior of the larvae was surprisingly different. The nauplii of *B. eburneus* and *B. amphitrite* occurred in maximal numbers at progressively greater depths during periods of low salinity, that is, at ebb tide. On the flood tide, however, the maximum numbers of nauplius stages I to III of these two species were found near the surface; the larval stages IV to VI were taken most abundantly at the 1.5-meter level. As for the third species, *B. improvisus*, the nauplius stages I to III were most abundant near the surface and the larval stages IV to cyprid were most plentiful at a depth of 3 meters (near the bottom), irrespective of the state of the tidal cycle. Near the head of the estuary, where tidal change in salinity is greatest, the number of larvae of *B. eburneus* and *B. amphitrite* fluctuate regularly in accordance with the stage of the tidal cycle. The larvae are most abundant at high water.

The weight of evidence suggests that retention of the larvae in numbers sufficient to ensure maintenance of the adult population within the river is made possible by a combination of the following biological and physical processes: (a) the high productivity of the barnacles, (b) the short planktonic life, (c) an upriver counter drift, and (d) a slow rate of tidal currents.

So far as could be established, distribution of the barnacle larvae was not correlated with changes in dissolved oxygen, *p*H, and turbidity of the water.

PROCEEDINGS AND ABSTRACTS FROM THE MEETINGS OF THE ELISHA MITCHELL SCIENTIFIC SOCIETY, 1960–1961

562d Meeting, October 11, 1960

H. E. LEHMAN (Zoology): *An Experimental Study of Hybrid Development in Sea Urchins.*

563d Meeting, November 15, 1960

DEPARTMENT OF GEOLOGY: A survey of current research in the Department of Geology. All members of the staff talked briefly about their current research. They were introduced by the departmental chairman, Dr. Roy L. Ingram.

564th Meeting, December 13, 1960

I. M. TAYLOR (Medicine): *Studies with the Isolated Perfused Rat Heart.*

W. F. LITTLE (Chemistry): *Organometallic Sandwich Compounds.*

565th Meeting, January 10, 1961

DEPARTMENT OF BACTERIOLOGY: Brief historical development of the department from its inception in 1929 to the present was presented by the chairman, Dr. D. A. MacPherson. The faculty personnel has been selected so that there is a specialist in each of the fields of bacteriology commonly associated with a medically oriented department. These fields include clinical bacteriology and infectious diseases, bacterial physiology, immunology, virology, biophysics, medical mycology, and microbial genetics.

Current research includes the bacteriology of pyelonephritis; oral complications of antibiotic therapy; toxic components of psittacosis viruses; isolation of viruses from the oral cavity; toxic cellular components of Group A streptococci; biophysical studies of macromolecules and viruses; metabolic aspects of bacterial ecology in host tissues; amino acid decarboxylases of oral lactobacilli; and genetic and cytologic studies on pathogenic yeasts. Brief presentations of their research were made by Drs. W. J. Cromartie, J. K. Spitznagel, D. G. Sharp, Abe Widra, and W. R. Straughn.

566th Meeting, February 14, 1961

DEPARTMENT OF STATISTICS: A survey of current research. Dr. Harold Hotelling reviewed the areas of research of each of the members of the department with emphasis on recent work and publication.

567th Meeting, March 14, 1961

RICHARD G. HISKEY (Chemistry): An important consideration in any synthetic approach to the hormone insulin is the introduction of the two interchain and the single interchain disulfide bonds. A method of synthesis of model unsymmetrical disulfides has been devised which involves the sequence:

$$RSH + (SCN)_2 \rightarrow R\text{-}S\text{-}S\text{-}CN \xrightarrow{R'SH} R\text{-}S\text{-}S\text{-}R'$$

The extension of this method to unsymmetrical disulfides containing cystine has been realized and thus provides a realistic route to the synthesis of "model insulin" peptides. Initial synthetic efforts directed toward the "20-membered ring" of insulin are described as well as experiments involving tetra- and pentapeptides containing two cysteine residues. A tentative route for the synthesis of insulin-type peptides was also presented.

568th Meeting, April 11, 1961

PSYCHOMETRIC LABORATORY: A survey of current research.

1. Lyle V. Jones, Director of the Laboratory, has co-authored a new text for the assessment of language deficit in aphasia, *i. e.*, impairment of communication due to brain damage. The test is designed to test differential impairment associated with visual or aural stimuli; with spoken, written, or gestural responses; and with the various symbol systems the subject is required to use.

2. R. Darrell Bock has completed a comprehensive psychometric analysis of the Rorschach and Holtzman ink-blot tests. The stability and dimensionality of scores from the two tests were compared by use of a multivariate generalization of the classical concept of test reliability.

3. Emir H. Shuford developed a method of measuring the utility of various commodities by asking subjects to choose between lotteries with different odds in which the commodities are prizes. An interesting property of this method is that the utilities obtained are measured on an absolute scale with subjective indifference as the zero point.

4. Thomas E. Jeffrey has been studying

preferences of factory workers for alternative incentive plans. The study makes use of a type of factorial paired comparisons design which makes possible the isolation and measurement of the effect on preference of various features of the incentive plans.

Similar projects are planned for the coming academic year. In addition, the staff of the Psychometric Laboratory will be responsible for graduate instruction in quantitative psychology.

569th Meeting, May 16, 1961

R. L. HUBBARD (Physics): Determination of Vibrational Force Fields in Polyatomic Molecules and an Application to the Deuteroethanes. (This dissertation earned for Mr. Hubbard the Coker Award for 1961).

ABSTRACTS OF PAPERS PRESENTED AT THE SPRING MEETING OF THE SOCIETY OF NORTH CAROLINA BACTERIOLOGISTS, MARCH 18, 1961, AT DURHAM, N. C.

QUANTITATIVE MEASUREMENTS OF THE DEGREE OF VIRUS AGGREGATION AND ITS EFFECT ON PLAQUE TITRATIONS. *G. J. Galasso and D. G. Sharp, University of North Carolina School of Medicine, Chapel Hill.* Several years ago Luria *et al.* showed that under especially favorable conditions each bacteriophage particle produced one plaque on a monolayer of sensitive cells. Although no one has yet made a similar demonstration for animal viruses, there is a widespread tendency to believe that each plaque is the result of infection by a single active virus particle. Our experiments show that virus particle suspensions prepared from infected cells by the usual methods are markedly aggregated. This evidence comes from electron-micrographs of virus particles sedimented from dilute suspension upon a smooth surface. Particles falling upon a cell monolayer for plaque titration presumably fall the same way, in groups of various sizes. Each group can produce only one plaque.

It is shown that the method of freeze-thaw, commonly used to release virus from the cell, yields virus which is still so aggregated as to make accurate counts impossible. This is also the case when virus suspensions are subjected to repeated sedimentation and resuspension, as is done in virus purification methods. The experiments presented show that three minutes' sonic vibration is sufficient to attain a high degree of virus dispersion. Enzyme treatment with trypsin-chymotrypsin is nearly as effective.

It has also been found that thoroughly dispersed virus will reaggregate on standing. Plaque titrations support the thesis that aggregated virus does not yield as high a titer as dispersed virus.

The experiments presented serve to indicate that the number of plaque-forming units is dependent on the total number of virus in the inoculum, the state of aggregation, and the fraction of those particles potentially able to produce plaques.

STUDIES ON THE PHYSIOLOGY AND INTERMEDIARY METABOLISM OF *Actinomyces Naeslundii*. *Bob Branch Buchanan and Leo Pine, Duke University School of Medicine, Durham.* *Actinomyces naeslundii* is a facultative anaerobe which carries out a homolactic acid fermentation of glucose in the absence of CO_2. If grown anaerobically in the presence of optimal concentrations of CO_2, the organism incorporates one equivalent of CO_2 and produces one equivalent of succinate, acetate, formate, and a smaller amount of lactate. Growth in the presence of CO_2 is at least ten times greater than in its absence. Data obtained from glucose-1, -2, and -6-C^{14} fermentations and the presence of aldolase activity in cell-free extracts indicate the Embden-Myerhof path to be the most significant route of glucose dissimilation. $C^{14}O_2$ is incorporated into the carboxyl groups of succinate in accordance with a Wood-Werkman type of reaction. Labeled aspartate and glutamate have been isolated from hydrolyzed cells incubated in the presence of $C^{14}O_2$. Glutamate was labeled exclusively in the alpha-carboxyl carbon. Malate and fumarate partially replace the requirement for CO_2; these acids effect a sixfold increment in cell yields and a twentyfold increment in total acetate and formate production over the CO_2-less control values. Succinate replaces CO_2 to the extent that it stimulates growth, but there is no increase in volatile acid production. Consequently, CO_2 is believed to have at least three functions: (1) It serves as a hydrogen acceptor in that it permits the oxidation of one of the two lactate molecules emanating from glucose to

acetate and formate; in effect, the second lactate molecule accepts this hydrogen and is converted to succinate with the incorporation of CO_2. As evidenced by increased cell yields, additional energy is made available to the organism in this process presumably by the formation of acetyl phosphate. (2) CO_2 provides a source of aspartic acid which, as shown by tracer experiments, does not enter the growing cell. The stimulation of growth in the presence of succinate would appear to be due to its providing a C_4-skeleton for synthesis of this amino acid. Malate and fumarate play dual roles in that they may accept hydrogen and presumably permit formation of acetyl phosphate and formate from pyruvate as well as provide a source of aspartate. (3) CO_2 has at least a third unknown function, for growth in its absence is never as great as that observed in its presence regardless of the CO_2-replacing factors which have been supplied.

THE ESSENTIALITY OF COBALT IN SYMBIOTIC NITROGEN FIXATION. *Shaukat Ahmet and Harold J. Evans, North Carolina State College, Raleigh.* Experiments were conducted to study the effect of small amounts of cobalt on the growth and composition of inoculated soybean plants without nitrogen and the uninoculated plants supplied with adequate fixed nitrogen. The culture solutions in deionized water contained all the known essential elements which were especially purified for removing traces of cobalt. The plants were grown in growth chambers with the necessary care and precautions to eliminate all possible contaminations. In the symbiotically grown plants the roots were inoculated with an effective strain of washed *Rhizobium japonicum*. The cobalt source from the seeds was further reduced by the removal of the cotyledons and by cutting back the shoot growth from time to time.

The results showed that 0.1 ppb of cobalt, when supplied in the nutrient solution, gave a vigorous and optimum plant growth, whereas plants without cobalt were greatly depressed in growth and showed symptoms similar to nitrogen deficiency. In the absence of supplied cobalt the various plant parts contained less total nitrogen and the leaves were very low in chlorophyll. The dry weight of root nodules was reduced and contained low hemoglobin and greatly reduced vitamin B_{12}. In the absence of cobalt supply, 25 per cent of the plants died, whereas plants receiving cobalt showed no mortality. The seed formation was eighty fold higher when cobalt had been supplied. The effect of cobalt was very specific, as none of the 12 elements tried on the basis of their possible removal in the purification could replace cobalt. The addition of cobalt could also cure the deficiency symptoms of the deficient plants and restore normal growth. Thus cobalt was essential for the growth of symbiotically grown plants.

However, further experiments have shown that cobalt is not essential when plants are supplied with adequate nitrogen and grown nonsymbiotically. Therefore, it is concluded that cobalt is most probably required for the growth of nodule bacteria, which fix nitrogen and thus promote the growth of soybean plants.

FACTORS INFLUENCING THE INDUCTION OF M-PHASE IN *Candida albicans*. *Abe Widra, University of North Carolina School of Medicine, Chapel Hill.* Among the factors enhancing M-phase growth in *C. albicans* are low temperature, anaerobiosis, and nutrient-poor media. It is possible, however, to produce M-phase in this organism at 37°C. on sabouraud dextrose agar (DIFCO) if sufficient inorganic phosphate is added. Potassium ion is also needed. In practice, a 1 per cent final concentration of dipotassium phosphate added to the commercial medium yields abundant M-phase. Strain differences in the species are noted. It is also possible to select smooth Y-phase microcolonies which breed true as they segregate out of the original inoculum of a single strain. The ability to form M-phase, given the proper environment, may therefore be used as a genetic marker. Varying the phosphate/dextrose ratio in sabouraud dextrose agar (DIFCO) resulted in the finding that an optimal ratio existed for cultivation of the Y-phase. The ratio of phosphate/reducing sugar is extremely high in corn meal agar (commonly used to produce M-phase) and low to the vanishing point in sabouraud dextrose agar. It is postulated that M-phase results from unbalanced growth (1) on nutrient-poor media due to failure to maintain intracellular -SH groups (Nickerson), and (2) on nutrient-poor *or* nutrient-rich media containing excess inorganic phosphate due to excess polyphosphate formation and the subsequent binding of intracellular magnesium. Magnesium ion will antagonize M-phase formation on phosphate-rich media at acid *p*H. This postulated mechanism of M-phase formation is consistent with (1) the staining pattern of intracellular polyphosphates, (2) the observed morphology and growth pattern on M-phase producing media, and (3) the observed effects of chelating

agents on the "metallo-" moiety in Nickerson's protein-disulfide reductase.

INGESTION OF LATEX PARTICLES BY CHICKEN FIBROBLASTS *in vitro*. *John R. Overman, Duke University School of Medicine, Durham.* The ingestion of polystyrene latex (PSL) particles measuring 0.264 μ in diameter has been demonstrated in chicken fibroblasts *in vitro*. PSL was added to cultures of chicken fibroblasts grown on carbon-collodion-coated steel grids, and these mounts removed at intervals of from one-half to 24 hours. The fixed and metal-shadowed whole-cell preparations revealed PSL in various stages of entry into these cells. PSL was found in invaginations of the cell wall and in vacuoles in the cytoplasm. Sections of cultured fibroblasts inoculated with PSL revealed PSL particles in cell vacuoles. The rate of particle uptake, however, was too low to be measured by the method used.

COMPARISON OF SONIC- AND PHENOL-PREPARED CELL WALLS OF *Chromobacterium Violaceum*. *J. M. Leatherwood, R. W. Wheat, and E. Rollins, Duke University School of Medicine, Durham.* Cell walls were obtained from fresh and acetone-dried *C. violaceum* cells by disruption in a 10 KC Raytheon Sonicator for 30 minutes, followed by extractions with molar NaCl, H_2O, trypsin, NaCl and H_2O. Phenol-prepared cell walls were obtained by extracting acetone-dried cells with 45 per cent and 90 per cent phenol at 65°C. for 30 minutes. Phenol-prepared cell walls contained 12 per cent nitrogen and 10 per cent DNA, sonic-prepared cell walls contained 7 per cent nitrogen and 1 per cent DNA, while glucosamine and muramic acid were present in approximately equimolar proportions and represented from 1.5 to 2.5 per cent of the cell-wall fractions. Sonic-prepared cell walls contained 2.2 per cent of fucosamine, a specific amino sugar of *C. violaceum* polysaccharide (Crumpton and Davies [1958] Biochem. J. 70: 729). However, phenol-prepared cell walls did not contain fucosamine. Sonication and extraction with NaCl, trypsin, and water were more effective in removing DNA and extraneous nitrogen, while phenol extraction was very effective in removing the specific (lipo) polysaccharide in preparing cell walls of *C. violaceum*.

ULTRA-STRUCTURE OF LONG FORMS OF *Mycobacterium Bovis* INDUCED BY SULFATE DEFICIENCY. *L. A. Wilson and J. K. Spitznagel, University of North Carolina School of Medicine, Chapel Hill.* Ultra-thin sectioning and electron microscopy have been used to study the fine structure of the long forms of *Mycobacterium bovis* produced by growth in a sulfate deficient medium. Modifications of various established techniques were needed to achieve acceptable sections of the cells due to the physiological composition of the genus *Mycobacterium*. Nucleases were used in an effort to substantiate the location and structure of the nucleus and cytoplasm in the cell.

The cell wall appeared to consist of two layers, a dense outer layer and a lighter inner one. Small dense granules that were uniformly dispersed in the cytoplasm of sulfate-replete cells were aggregated in "lacelike" patterns in the deficient organisms. The fibrous material of the nucleus appeared to be compressed into very dense material by large vacuoles in the long forms. Round or oval "mitochondrial" structures were observed with a limiting membrane as well as double membranous structures that did not have any distinctive shape. Large electron-dense granules were shown to have a "honey-combed" structure, to be located terminally in normal cells, and to have a more central location in the long forms. The location may be an indication of the mode of growth of the long forms of *M. bovis*.

ABSTRACTS OF PAPERS PRESENTED AT THE ANNUAL MEETING OF THE NORTH CAROLINA PSYCHOLOGICAL ASSOCIATION, MAY 6, 1961, AT RALEIGH, N. C.

THE EVIDENCE FOR ESP. *J. G. Pratt, Duke University.* The key word in the topic before the panel is *evidence*. The question of the occurrence of ESP has long been investigated by laboratory methods, particularly those of experimental psychology. Questions about the credibility of the evidence were raised more frequently at an earlier stage of the research and were answered by specific controls introduced into the experimental procedures. This period of the "ESP controversy" ended with the publication of the book *Extrasensory Per-*

ception after Sixty Years by Rhine *et al.* in 1940.

During the twenty years since that time, the strength of the experimental evidence for ESP has greatly increased. This period has been primarily characterized, however, by an increase in knowledge *about* ESP, and a rational picture of the function and its place in the general psychology of personality has started to take shape. This organized body of knowledge about ESP has become strong confirmatory evidence for the reality of the phenomenon.

The accumulated evidence has had a strong impact upon scientists outside of parapsychology who have examined the facts. Some critics have frankly admitted that there is no escaping the conclusions except on the assumption of a widespread hoax involving academic experimenters. Others have frankly admitted that their rejection is on the basis of prejudice. A number of opinion surveys among psychologists reveal that a majority of those with adverse opinions have not examined the research reports. At the same time, there has been a steady increase in the percentage of those who indicated that they considered ESP as either established or a likely possibility.

A BRIEF METHODOLOGICAL ANALYSIS OF EXTRASENSORY PERCEPTION RESEARCH. *John Oliver Cook, North Carolina State College.* The viewpoint propounded in this paper is that, as a result of a misunderstanding concerning the meaning of the ESP hypothesis, the bulk of the data gathered in ESP research is irrelevent. A sensible ESP hypothesis would not assert that improbably high scores in a guessing situation are impossible to obtain by chance. Hence, it is concluded that the thesis that an ESP ability exists in people must mean that, in a guessing situation, scores higher than the chance mean occur with a frequency greater than the chance expectation. But ESP research has not been directed toward testing this hypothesis.

CAN SCIENCE AND ESP LIVE TOGETHER? *Harold G. McCurdy, University of North Carolina.* The author accepts ESP on the basis of personal experience and the anecdotal literature. The statistical and experimental literature does not pay enough attention to the equality of personal significance in these experiences and it cannot, in his opinion, convince the skeptical. There might be some value, however, in more systematic exploration of certain hypotheses about the conditions favorable for ESP, as, for example, emotional states, brain pathology, and the psychotherapeutic situation. Bits of evidence bearing on these hypotheses are presented. The paper concludes with the observation that certain conditions favoring ESP may decrease interest in the phenomena as such, as in the case of mystics and Christian saints, and thus put the best potential subjects beyond the reach of experimental science.

THE RATIONAL UNIVERSITY. *Howard G. Miller. North Carolina State College.* In the wide range of educational problems dealt with in the modern university, there is generally little or no attempt to employ orderly and rational procedures in their solutions. Included in such problems are those related to curriculum design, course and class scheduling, teaching methods, student incentives, and faculty incentives. This failure is in marked contrast to advanced practice in modern industry and government, where, as a result of the development of modern organization theory and certain mathematical and analytic techniques, very significant progress has been made in the effective rational solution of similar problems.

In particular, the university does not deal with the teaching functions in a rational, orderly manner. Underlying this failure is the absence of a general theory of education which takes into account the chief factors involved in the educational process. Without such a theory it is impossible to formulate hypotheses and to make deductions which enable us to deal with educational problems effectively. We are inclined to wander aimlessly in our search for better means, if we are stimulated to search at all.

A first example of our failure to follow rational procedures is that of the method employed in teaching. We are accustomed to choose our methods on the basis of tradition, or habit, or skill, or convenience, but almost never because a particular method has been discovered to produce a certain educational objective effectively. In fact we simply do not possess the knowledge to make such decisions. The absence of a general theory has been a fundamental contributor to the failure of experimentation in teaching methods to produce a consistent body of empirical data which would aid in the making of such decisions.

A systems approach to the problem of the construction of educational theory seems to be a productive approach to this problem. Using such an approach, the teaching resources become components or elements in the system

along with the student or learner. The measure of the effectiveness of the system is the amount learned by the student. With a theoretical framework derived from such a scheme more productive data generation could develop rapidly.

A second educational problem which illustrates the failure to employ rational procedures is in the scheduling of student work and class activities. The traditional system calls for the scheduling of five or six courses simultaneously over a period of sixteen weeks in length. The sole justification for such an arrangement seems to be traditional and administrative convenience. Certainly the work pattern of students is not a matter of indifference, since effective work patterns will greatly enhance student performance. In any scheduling arrangement issues such as the optimal number of courses to be taken simultaneously, the effect of schedules on such methods as independent study, the interaction effects of various courses and other educational factors should be considered. We do not now have a theory or any data which will enable us to schedule student work efficiently, nor are we ever led to ask the right questions.

There are many other problems in the university requiring effective rational analysis. Until we develop appropriate theoretical structures and attempt rational solutions to these problems, university education will continue in its present unproductive ways.

THE REPTILES AND AMPHIBIANS OF NORTH CAROLINA: A Preliminary Check List and Bibliography

CHARLES E. DEPOE, JOHN B. FUNDERBURG, JR., AND THOMAS L. QUAY
Department of Zoology, North Carolina State College, Raleigh, N. C.

During the period from 1907 to 1926, the late Dr. Clement S. Brimley, dean of North Carolina herpetologists, published five check lists to the reptiles and amphibians of North Carolina in the *Journal of the Elisha Mitchell Scientific Society*. In addition, he also published numerous other papers on the herpetology of the state in this journal as well as in many others. He culminated his herpetological writings with his annotated list "Amphibians and Reptiles of North Carolina" (Brimley, 1943), which was published as a series in *Carolina Tips* from 1939 to 1943. In this check list he recorded the counties from which each form was known at that time, but the list was neither exhaustive nor complete.

Since 1943 there has been a tremendous amount of research done on the herpetofauna of North Carolina, especially in the mountain region. However, no further compilation has been made of all the forms occurring in the state. Some county lists, such as those for New Hanover (Funderburg, 1955) and Pitt (Eaton, 1953), have been published but such accounts are few. Furthermore, they do not usually include a review of all of the orders. Other general papers, such as Roberson and Tyson's (1950) "Herpetological Notes from Eastern North Carolina," cover many of the species but are also incomplete in that they do not list all forms from any one county or area.

North Carolina is ideally suited to support a wide variety of reptiles and amphibians. Physiographically speaking, the state can be divided into three main regions: the Coastal Plain, the Piedmont, and the mountains. These three areas not only provide an abundant range of habitat types, but also provide a wide variation in climatic conditions.

The coastal plain region contains approximately 21,000 square miles, roughly two-fifths of the state's total area, and extends from the Atlantic Ocean westward to the Fall Line. Its maximum width is about 125 miles. The elevation rises very gradually from sea level along the eastern side to over 400 feet in some localities along the Fall Line. Much of the eastern area is, however, below twenty-five feet in altitude and is composed largely of swamps, bays, marshes, and other lowlands. The inland portion, while still relatively flat, consists of gently rolling hills and sandy soils.

The piedmont region extends westward from the Fall Line nearly 200 miles to an altitude of about 1,500 feet at the base of the Blue Ridge Mountains. It encompasses about 22,000 square miles and is characterized by a gently rolling topography occasionally broken by steep ranges of high hills, such as the Uwharrie Mountains, King's Mountain, and the South Mountains. Generally speaking, the predominant soil throughout is clay. The entire Piedmont is

broadly dissected by intermittent streams, most of which eventually drain into large rivers which transect the Coastal Plain. During the past decade, the drainage pattern has been much interrupted by the construction of numerous farm ponds which tend to change the volume and regulate seasonal fluctuations in these streams.

The mountain region rises sharply from the Piedmont to form, on the eastern side, the Blue Ridge Mountains (with an elevation between 3,000 and 4,000 feet) and, on the western border of the state, the Great Smoky Mountains (which range to above 6,000 feet). Between these large ranges are various cross ridges and prominent valleys. The entire mountain region is about 200 miles in length and varies between 15 and 50 miles in width. It covers approximately 6,000 square miles.

The wide diversity of geographic situations also produces highly varied climatic conditions (for example, Smith Island, at the mouth of the Cape Fear River, is a subtropical area). The general distribution of our herpetofauna is closely associated with this geographic and climatic diversity. Many forms which are typically more southern extend northward in the Coastal Plain. This distributional pattern is subject, however, to marked modification by man's activities. The clearing of wooded areas and the draining of marshlands, as well as the pollution of streams and rivers by sewage and industrial wastes, are examples of the many ways by which forms which were once common have been exterminated. In contrast, the construction of farm ponds has created an entirely new habitat in many areas and this factor has permitted an extension of range by many forms.

The herpetofauna of North Carolina is composed of 158 species and subspecies recorded, and another 11 forms which may possibly occur but which currently appear doubtful. These are divided among the five orders as follows:

Crocodilia (alligators)	1
Chelonia (turtles)	17 + 3 possible
Squamata (lizards and snakes)	
Suborder Lacertilia (lizards)	11
Suborder Ophidia (snakes)	46 + 3 possible
Urodela (salamanders)	53 + 4 possible
Anura (frogs and toads)	30 + 1 possible

The annotated list of all species and subspecies known from North Carolina is presented below. Each species is described in two sets of entries. The common and scientific names are given first and are in accordance with *A Field Guide to Reptiles and Amphibians* (Conant, 1958), the largest and most up-to-date compilation of all forms known from the eastern United States. Following the scientific name is the known range of the species in the state.

The range of a form is given, in most cases, as that general section of the state in which the form may be expected to occur. Exceptions have been made where the known ranges in adjacent states would aid in understanding the situation in North Carolina. Ranges have been established by a compilation of: (1) the counties from which the form has been reported (a card file containing all published county records available for each species is in the senior author's possession), (2) known ranges outside North Carolina, (3) previously reported ranges within North Carolina, and (4) a consideration of general distribution patterns within the state. Forms which have been reported but apparently are in error, or which should be expected but have not been reported, have been enclosed in brackets.

Lower Coastal Plain refers to the eastern and southern portions of the Coastal Plain characterized by wet marshes, swamps, and other lowland areas. The *upper Coastal Plain* is the higher and drier region between the lower Coastal Plain and the Fall Line. The *lower Piedmont* is the eastern portion of the piedmont plateau in which the topography maintains a gently rolling aspect but remains comparatively level. The *upper Piedmont* is the western area of the Piedmont which assumes a more dissected topography as the mountains are reached. This is the "foothill" region. *Mountains* refers to the mountainous regions in the western part of the state.

ANNOTATED LIST

Order Crocodila: Alligators and Crocodiles

This order is represented in North Carolina by only one form, the American alligator. Very abundant at one time, the encroachment of man upon its habitat, extensive market hunting, and indiscriminate shooting have greatly reduced its numbers. The present decline in demand for its hide, however, has allowed the continued existence of small populations in a few restricted areas in the lower Coastal Plain.

American Alligator, *Alligator mississipiensis*—lower Coastal Plain south of Albemarle Sound.

Order Chelonia: Turtles

For the most part, the turtles present few problems aside from the need for records from

more areas, particularly the western part of the state. However, the distribution of *Malaclemys terrapin* along the coast and the *Pseudemys* complex are both in need of further work. In all work with turtle identification, the wide range of possible variation within a single form must be kept constantly in mind.

Common Snapping Turtle, *Chelydra serpentina serpentina*—statewide.

Stinkpot, *Sternothaerus odoratus*—statewide.

Eastern Mud Turtle, *Kinosternon subrubrum subrubrum*—lower Piedmont and the Coastal Plain.

Spotted Turtle, *Clemmys guttata*—Coastal Plain and lower Piedmont.

Bog Turtle, *Clemmys muhlenbergi*—mountains and upper Piedmont.

Eastern Box Turtle, *Terrapene carolina carolina*—statewide.

Northern Diamondback Terrapin, *Malaclemys terrapin terrapin*—coastal marshes south to Cape Hatteras.

Carolina Diamondback Terrapin, *Malaclemys terrapin centrata*—coastal marshes north to Cape Hatteras, intergrading with *M. t. terrapin* where their ranges meet.

Eastern Painted Turtle, *Chrysemys picta picta*—Piedmont and Coastal Plain.

Yellow-Bellied Turtle, *Pseudemys scripta scripta*—Coastal Plain and lower Piedmont.

River Cooter, *Pseudemys concinna concinna*—Piedmont, intergrading with *P. f. floridana* along the Fall Line.

Florida Cooter, *Pseudemys floridana floridana*—Coastal Plain. (See *Pseudemys concinna*.)

[Red-Bellied Turtle, *Pseudemys rubiventris*—the Virginia Coastal Plain and northward. Possibly in the extreme northern Coastal Plain of North Carolina.]

Eastern Chicken Turtle, *Deirochelys reticularia reticularia*—Coastal Plain south and west of Carteret County.

Atlantic Green Turtle, *Chelonia mydas mydas*—a summer resident occasionally found along the coast and in the salt-water sounds. Formerly abundant in the vicinity of Beaufort.

[Atlantic Hawksbill Turtle, *Eretmochelys imbricata imbricata*—a possible straggler along the North Carolina coast since, like the other marine turtles, it ranges far north of the state. However, there is no valid record from North Carolina.]

Atlantic Loggerhead Turtle, *Caretta caretta caretta*—a summer resident, common in the salt-water sounds and off the coast. Breeds along the ocean beaches, at least south of Beaufort (Carteret County).

Atlantic Ridley, *Lepidochelys kempi*—an occasional straggler along the coast. Formerly common in the vicinity of Beaufort (Carteret County).

Atlantic Leatherback Turtle, *Dermochelys coriacea coriacea*—an occasional straggler off the coast. Prefers deeper seas more than the other marine turtles.

[Gulf Coast Softshell Turtle, *Trionyx spinifer asper*—Coastal Plain.]

Order Squamata: Lizards and Snakes

Suborder Lacertilia: Lizards

The major problems which need investigation among lizards are the distribution of *Eumeces a. anthracinus* and *E. a. pluvialis* in the mountains; the distribution of *Eumeces fasciatus, E. laticeps,* and *E. inexpectatus* throughout the state; and the establishment of the northward limits of *Sceloporus u. undulatus,* since it may enter our Coastal Plain. A better knowledge of the relative abundance of all forms in various habitats would also be valuable.

Green Anole, *Anolis carolinensis carolinensis*—statewide.

Northern Fence Lizard, *Sceloporus undulatus hyacinthus*—statewide.

Six-Lined Racerunner, *Cnemidophorus sexlineatus sexlineatus*—extreme southeastern Coastal Plain only (see next race).

Six-Lined Racerunner, *Cnemidophorus sexlineatus pauciporus*—Piedmont and mountains. Intergrades with *C. s. sexlineatus* throughout the Coastal Plain except in extreme southeastern North Carolina.

Ground Skink, *Lygosoma laterale*—Piedmont and Coastal Plain.

Five-Lined Skink, *Eumeces fasciatus*—statewide.

Broad-Headed Skink, *Eumeces laticeps*—statewide.

Southeastern Five-Lined Skink, *Eumeces inexpectatus*—Coastal Plain, plus scattered areas in the Piedmont and mountains.

Southern Coal Skink, *Eumeces anthracinus anthracinus*—mountains of southwestern North Carolina.

Eastern Glass Lizard, *Ophisaurus ventralis*—statewide.

Eastern Slender Glass Lizard, *Ophisaurus attenuatus longicaudus*—lower Piedmont and the Coastal Plain.

Suborder Ophidia: Snakes

There are several problems within this group. Among these are the present distribu-

tion of *Crotalus h. horridus* and *C. h. atricaudatus;* the relationship between *Natrix s. sipedon* and *C. h. fasciata,* since the two forms are subspecies and should therefore be expected to intergrade where their ranges meet (but this does not appear to be so); the distribution of *Storeria d. dekayi* and *D. d. wrightorum;* and the status and distribution of the subspecies of *Lampropeltis doliata.*

Brown Water Snake, *Natrix taxispilota*—Coastal Plain.

Red-Bellied Water Snake, *Natrix erythrogaster erythrogaster*—Coastal Plain.

Northern Water Snake, *Natrix sipedon sipedon*—mountains, Piedmont, and possibly to the Atlantic Coast north of Albemarle Sound. May intergrade with *N. s. fasciata* where their ranges are contiguous.

Midland Water Snake, *Natrix sipedon pleuralis*—described by Cope in 1892 from Summerville (Harnett County), N. C. However, Stejneger and Barbour (1939) have said that this record was probably from Summerville, South Carolina, which would put it within the normal range of this form as it is known today. The only other record from the state is one specimen from Tusquitee, Clay County (Cliburn, 1958).

Banded Water Snake, *Natrix sipedon fasciata*—Coastal Plain, at least south of Albemarle Sound.

Queen Snake, *Natrix septemvittata*—statewide, except lower Coastal Plain.

Glossy Water Snake, *Natrix rigida*—lower Coastal Plain.

Carolina Swampsnake, *Seminatrix pygaea paludis*—lower Coastal Plain south of the Neuse River.

Brown Snake, *Storeria dekayi*—statewide, with probably a wide zone of intergradation between *S. d. dekayi* and *S. d. wrightorum* (See Conant, 1958).

Northern Red-Bellied Snake, *Storeria occipitomaculata occipitomaculata*—statewide.

Eastern Garter Snake, *Thamnophis sirtalis sirtalis*—statewide.

Eastern Ribbon Snake, *Thamnophis sauritus sauritus*—statewide.

Rough Earth Snake, *Haldea striatula*—Piedmont and Coastal Plain.

Eastern Earth Snake, *Haldea valeriae valeriae*—statewide.

Eastern Hognose Snake, *Heterodon platyrhinos*—statewide.

Southern Hognose Snake, *Heterodon simus*—Coastal Plain.

Yellow-Lipped Snake, *Rhadinea flavilata*—lower Coastal Plain south of the Neuse River.

Southern Ringneck Snake, *Diadophis punctatus punctatus*—Coastal Plain and lower Piedmont.

Northern Ringneck Snake, *Diadophis punctatus edwardsi*—mountains and upper Piedmont.

Eastern Worm Snake, *Carphophis amoenus amoenus*—statewide.

Rainbow Snake, *Abastor erythrogrammus*—Coastal Plain.

Eastern Mud Snake, *Farancia abacura abacura*—Coastal Plain.

Northern Black Racer, *Coluber constrictor constrictor*—statewide, with possible exception of extreme southeastern North Carolina.

[Southern Black Racer, *Coluber constrictor priapus*—racers from extreme southeastern North Carolina probably belong to this race.]

Eastern Coachwhip, *Masticophis flagellum flagellum*—southern half of the Coastal Plain.

Rough Green Snake, *Opheodrys aestivus*—statewide.

Eastern Smooth Green Snake, *Opheodrys vernalis vernalis*—scattered areas of the mountains north of the French Broad River.

Corn Snake, *Elaphe guttata guttata*—statewide.

Black Rat Snake, *Elaphe obsoleta obsoleta*—statewide, with the exception of the lower Coastal Plain. Intergrades with *E. o. quadrivittata* in the latter area.

Yellow Rat Snake, *Elaphe obsoleta quadrivittata*—lower Coastal Plain south of Albemarle Sound, ranging north to Hatteras Island on the other banks.

[Gray Rat Snake, *Elaphe obsoleta spiloides*—probably not in North Carolina; in the Coastal Plain of Georgia and southward.]

Northern Pine Snake, *Pituophis melanoleucus melanoleucus*—possibily statewide.

[Florida Pine Snake, *Pituophis melanoleucus mugitus*—this form may possibly occur in southeastern North Carolina (Wright and Wright, 1957).]

Eastern Kingsnake, *Lampropeltis getulus getulus*—statewide.

Scarlet Kingsnake, *Lampropeltis doliata doliata*—Coastal Plain and Piedmont.

Eastern Milk Snake, *Lampropeltis doliata triangulum*—mountains.

Coastal Plain Milk Snake, *Lampropeltis doliata temporalis*—northern Piedmont and Coastal Plain.

Mole Snake, *Lampropeltis calligaster rhombomaculata*—Piedmont and Coastal Plain.

Scarlet Snake, *Cemophora coccinea*—Coastal Plain and Piedmont.

Southeastern Crowned Snake, *Tantilla coronata coronata*—Coastal Plain and lower Pied-

mont. Intergrades with *T. c. mitrifer* at the juncture of their ranges.

Appalachian Crowned Snake, *Tantilla coronata mitrifer*—southern mountains.

Eastern Coral Snake, *Micrurus fulvius fulvius*—southern half of the Coastal Plain.

Southern Copperhead, *Agkistrodon contortrix contortrix*—Coastal Plain and lower Piedmont. May intergrade with *A. c. mokeson* in restricted areas.

Northern Copperhead, *Agkistrodon contortrix mokeson*—mountains and upper Piedmont.

Eastern Cottonmouth, *Agkistrodon piscivorus piscivorus*—Coastal Plain.

Carolina Pigmy Rattlesnake, *Sistrurus miliarius miliarius*—Coastal Plain south of Albemarle Sound.

Timber Rattlesnake, *Crotalus horridus horridus*—almost statewide, replacing or intergrading with *C. h. atricaudutus* in the Coastal Plain.

Canebrake Rattlesnake, *Crotalus horridus atricaudatus*—lower Coastal Plain.

Eastern Diamondback Rattlesnake, *Crotalus adamanteus*—Coastal Plain.

Order Urodela: Salamanders

The Urodela, or salamanders, comprise the largest amphibian order in North Carolina, and the North Carolina mountains are the center of distribution for many forms. There is, therefore, a great deal of literature on this group. Nevertheless, the status of many species and subspecies is still unknown.

Many of the forms give difficulty because of changes in nomenclature. As knowledge of this group has accumulated, groups once designated as species have been reduced to subspecific status, and vice versa. In many forms taxonomic studies are still in progress.

Hellbender, *Cryptobranchus alleganiensis alleganiensis*—mountains.

Neuse River Waterdog, *Necturus lewisi*—Neuse and Tar River systems.

Dwarf Water Dog, *Necturus punctatus punctatus*—streams of the southern Coastal Plain.

Greater Siren, *Siren lacertina*—lower Coastal Plain.

Two-Toed Amphiuma, *Amphiuma means means*—Coastal Plain.

Frosted Flatwoods Salamander, *Ambystoma cingulatum cingulatum*—reported only from Brunswick County, but may occupy other areas in the southern Coastal Plain.

[Jefferson Salamander, *Ambystoma jeffersonianum*—recorded from Roan Mountain (Mitchell County) but probably in error.]

Mabee's Salamander, *Ambystoma mabeei*—lowlands of the Cape Fear River system.

[Small-Mouthed Salamander, *Ambystoma texanum*—although reported from Harnett and Rutherford counties, this salamander is generally considered to be limited to areas west of the Appalachians. Furthermore, no records have been made since 1929, which makes the above reports dubious.]

Mole Salamander, *Ambystoma talpoideum*—reported from only Henderson and Transylvania counties.

Marbled Salamander, *Ambystoma opacum*—Piedmont and Coastal Plain.

Spotted Salamander, *Ambystoma maculatum*—statewide.

Eastern Tiger Salamander, *Ambystoma tigrinum tigrinum*—Coastal Plain.

Red-Spotted Newt, *Diemictylus viridescens viridescens*—statewide, except for the southeastern Coastal Plain, where it intergrades with *D. v. dorsalis*.

Broken-Stripe Newt, *Diemictylus viridescens dorsalis*—southern half of the Coastal Plain.

Northern Dusky Salamander, *Desmognathus fuscus fuscus*—statewide, except for the lower Coastal Plain.

Southern Dusky Salamander, *Desmognathus fuscus auriculatus*—Coastal Plain.

Blue Ridge Mountain Salamander, *Desmognathus ocrophaeus carolinensis*—mountains.

Cliffside Salamander, *Desmognathus perlapsus*—recorded from only Cold Mountain (Transylvania County) at an altitude of 4,400 feet.

Appalachian Seal Salamander, *Desmognathus monticola monticola*—mountains, below 5,000 feet.

Black-Bellied Salamander, *Desmognathus quadramaculatus*—mountains.

Pigmy Salamander, *Desmognathus wrighti*—mountains, above 3,500 feet.

Cherokee Salamander, *Desmognathus aeneus*—known only from the type locality near Peachtree in Cherokee County.

Northern Shovel-Nosed Salamander, *Leurognathus marmoratus marmoratus*—mountains northeast of the French Broad River, between 2,500 and 4,000 feet altitude.

Golden Shovel-Nosed Salamander, *Leurognathus marmoratus aureatus*—headwaters of the Tallulah and Chattahoochee rivers in northeastern Georgia and southwestern North Carolina.

Husky Shovel-Nosed Salamander, *Leurognathus marmoratus roboratus*—Chattoooga River

in North Carolina, South Carolina, and Georgia.

Southern Shovel-Nosed Salamander, *Leurognathus marmoratus intermedius*—In the Pigeon and Tuchasegee River drainages of the southern mountains, between 1,500 and 5,400 feet altitude.

Black Shovel-Nosed Salamander, *Leurognathus marmoratus melanius*—mountains of southwestern North Carolina.

Red-Backed Salamander, *Plethodon cinereus cinereus*—statewide.

[Zigzag Salamander, *Plethodon dorsalis*—recorded from only Asheville (Buncombe County), but probably in error.]

Southern Ravine Salamander, *Plethodon richmondi popei*—mountains of northwestern North Carolina.

Weller's Salamander, *Plethodon welleri welleri*—restricted to higher altitudes on Grandfather Mountain, North Carolina.

Spot-Bellied Salamander, *Plethodon welleri ventromaculatum*—mountains of northwestern North Carolina, except Grandfather Mountain.

Slimy Salamander, *Plethodon glutinosus glutinosus*—statewide.

Carolina Slimy Salamander, *Plethodon glutinosus chlorobryonis*—lower Coastal Plain, however, there is some doubt as to the validity of this race.

Yonahlossee Salamander, *Plethodon yonahlossee*—mountains east and north of the French Broad River, from 2,500 to 5,700 feet altitude.

Red-Cheeked Salamander, *Plethodon jordani jordani*—the Great Smoky Mountains along the North Carolina–Tennessee border in the region of Swain and Haywood counties at elevations of about 2,500 feet.

Red-Legged Salamander, *Plethodon jordani shermani*—Nantahala Mountains in southwestern North Carolina above 2,000 feet in altitude.

Metcalf's Salamander, *Plethodon jordani metcalfi*—mountains, south to Jackson and Swain counties, between 3,500 and 5,800 feet in altitude.

Highlands Salamander, *Plethodon jordani melaventris*—mountains south of Buncombe County at the altitudes between 2,500 and 4,500 feet.

Teyahalee Salamander, *Plethodon jordani teyahalee*—restricted to the Snowbird Mountains in southwestern North Carolina at altitudes between 4,000 and 4,500 feet.

Four-Toed Salamander, *Hemidactylium scutatum*—recorded from only Durham County (Duke Forest).

Many-Lined Salamander, *Stereochilus marginatus*—Coastal Plain.

[Northern Spring Salamander, *Gyrinophilus porphyrictus porphyrictus*—west of the Appalachian Mountains. Recorded from Roan Mountain (Mitchell County) in 1895 but apparently in error.]

Blue Ridge Spring Salamander, *Gyrinophilus danielsi danielsi*—from the vicinity of Grandfather Mountain to and including the Great Smoky Mountains.

Mount Mitchell Spring Salamander, *Gyrinophilus danielsi polystictus*—recorded only from Mount Mitchell and Blackstock Knob Mountain at altitudes above 4,500 feet.

Carolina Spring Salamander, *Gyrinophilus danielsi dunni*—mountains below 3,500 feet in altitude.

Eastern Mud Salamander, *Pseudotriton montanus montanus*—statewide.

Northern Red Salamander, *Pseudotriton ruber ruber*—Piedmont and Coastal Plain.

Blue Ridge Red Salamander, *Pseudotriton ruber nitidus*—mountains north and east of the French Broad River.

Black-Chinned Red Salamander, *Pseudotriton ruber schencki*—mountains south and west of the French Broad River.

Green Salamander, *Aneides aeneus*—mountains.

Southern Two-Lined Salamander, *Eurycea bislineata cirrigera*—Piedmont and Coastal Plain.

Blue Ridge Two-Lined Salamander, *Eurycea bislineata wilderae*—Blue Ridge Mountains.

Long-Tailed Salamander, *Eurycea longicauda longicauda*—mountains.

Three-Lined Salamander, *Eurycea longicauda guttolineata*—statewide.

Dwarf Salamander, *Manculus quadridigitatus*—Coastal Plain south of Albemarle Sound.

Order Anura: Frogs and Toads

Eastern Spadefoot Toad, *Scaphiopus holbrooki*—possibly statewide, but reported only from the Coastal Plain and two mountain counties.

American Toad, *Bufo americanus americanus*—Piedmont and mountains.

Southern Toad, *Bufo terrestris*—Coastal Plain.

Fowler's Toad, *Bufo woodhousei fowleri*—statewide.

Oak Toad, *Bufo quercicus*—Coastal Plain.

Southern Cricket Frog, *Acris gryllus gryllus*—Coastal Plain.

Northern Cricket Frog, *Acris crepitans crepitans*—Piedmont and mountains.

Northern Spring Peeper, *Hyla crucifer crucifer*—statewide.

Green Treefrog, *Hyla cinerea cinerea*—Coastal Plain.

Pine Barrens Treefrog, *Hyla andersoni*—known only from Moore and Richmond counties in North Carolina.

Pine Woods Treefrog, *Hyla femoralis*—Coastal Plain.

Squirrel Treefrog, *Hyla squirrella*—Coastal Plain.

Eastern Gray Treefrog, *Hyla versicolor versicolor*—statewide.

Barking Treefrog, *Hyla gratiosa*—lower Coastal Plain south of Beaufort County.

Little Grass Frog, *Hyla ocularis*—Coastal Plain.

Upland Chorus Frog, *Pseudacris triseriata feriarum*—statewide, except lower Coastal Plain.

Southern Chorus Frog, *Pseudacris nigrita nigrita*—Coastal Plain.

Brimley's Chorus Frog, *Pseudacris brimleyi*—Coastal Plain.

Mountain Chorus Frog, *Pseudacris brachyphona*—reported only from Cherokee County.

Ornate Chorus Frog, *Pseudacris ornata*—lower Coastal Plain south of Beaufort County.

Eastern Narrow-Mouthed Toad, *Microhyla carolinensis*—statewide.

Bullfrog, *Rana catesbeiana*—statewide.

River Frog, *Rana heckscheri*—southeastern Coastal Plain.

Carpenter Frog, *Rana virgatipes*—Coastal Plain.

Bronze Frog, *Rana clamitans clamitans*—Coastal Plain.

Green Frog, *Rana clamitans melanota*—mountains and Piedmont.

[Northern Leopard Frog, *Rana pipiens pipiens*—possibly present in the mountains and upper Piedmont. Dubiously recorded from Edgecombe and Durham counties.]

Southern Leopard Frog, *Rana pipiens sphenocephala*—Coastal Plain and Piedmont.

Pickerel Frog, *Rana palustris*—statewide.

Wood Frog, *Rana sylvatica*—mountains.

Carolina Gopher Frog, *Rana areolata capito*—lower Coastal Plain.

BIBLIOGRAPHY

BAILEY, JOSEPH R. 1937. Notes on the Plethodont Salamanders of the Southeastern United States. Univ. Mich. Occ. Papers Mus. Zool. No. 364.

BAIRD, SPENCER F., AND CHARLES GIRARD. 1853. Catalogue of North American Reptiles in the Museum of the Smithsonian Institute. Part I. Serpents. Publ. Smithsonian Inst.

BARBOUR, THOMAS R. 1937. Herpetological notes regarding *Diadophis punctatus*. Proc. New England Zool. Club **7**: 7–13.

———. 1943. A new water snake from North Carolina. Proc. New England Zool. Club **22**: 1–2.

BARBOUR, THOMAS, AND W. L. ENGELS. 1942. Two interesting new snakes. Proc. New England Zool. Club **20**: 101–104.

BISHOP, SHERMAN C. 1924. Notes on salamanders. N. Y. State Mus. Bull. **253**: 87–102.

———. 1925. Records of some salamanders from North Carolina and Pennsylvania. Copeia **139**: 9–12.

———. 1928. Notes on some amphibians and reptiles from the southeastern states with a description of a new salamander from North Carolina. Jour. Elisha Mitchell Sci. Soc. **43**: 153–170.

———. 1941. Notes on Salamanders with Descriptions of Several New Forms. Univ. Mich. Occ. Papers Mus. Zool. No. 451.

———. 1943. Handbook of Salamanders. Comstock Publ. Co.: Ithaca, N. Y.

BLAIR, FRANK W., ALBERT P. BLAIR, FRED R. CAGLE, AND GEORGE A. MOORE. 1957. Vertebrates of the United States. McGraw-Hill Book Co.: New York.

BLANCHARD, FRANK N. 1920a. Three New Snakes of the Genus *Lampropeltis*. Univ. Mich. Occ. Papers Mus. Zool. No. 81.

———. 1920b. A Synopsis of the King Snakes: Genus *Lampropeltis* Fitzinger. Univ. Mich. Occ. Papers Mus. Zool. No. 87.

BRADY, MAURICE. 1927. Notes on the reptiles and amphibians of the Dismal Swamp. Copeia **162**: 26–29.

BRANDT, B. B. 1933. An extension of the range of *Hyla gratiosa* Le Conte. Copeia **1933**: 39.

———. 1934. The Salientia of Beaufort County, North Carolina. Unpublished master's thesis, Duke University, Durham, N. C.

———. 1936a. Parasites of certain North Carolina Salientia. Ecol. Monographs **6**: 491–532.

———. 1936b. The frogs and toads of eastern North Carolina. Copeia **1936**: 215–223.

BRANDT, B. B. AND C. F. WALKER. 1933. A New Species of *Pseudacris* from the Southeastern United States. Univ. Mich. Occ. Papers Mus. Zool. No. 272.

BREDER, CHARLES M., JR. AND R. B. BREDER. 1923. A list of the fishes, amphibians and reptiles collected in Ashe County, North Carolina. Zoologica **4**: 1–23.

BRIMLEY, C. S. 1895a. List of snakes observed at Raleigh, N. C. Am. Nat. **29**: 56–57.

———. 1895b. Habits of *Heterodon platyrhinos* at Raleigh, N. C. Am. Nat. **29**: 75.

———. 1896. Batrachia found at Raleigh, N. C. Am. Nat. **30**: 500–501.

———. 1904. The box tortoises of southeastern North America. Jour. Elisha Mitchell Sci. Soc. **20**: 27–34.

———. 1905. Notes on the scutellation of the red king snake, *Ophiobolus doliatus coccineus* Schlegel. Jour. Elisha Mitchell Sci. Soc. **21**: 145–148.

———. 1907a. Notes on some turtles of the genus *Pseudemys*. Jour. Elisha Mitchell Sci. Soc. **23**: 76–84.

———. 1907b. Artificial key to the species of snakes and lizards which are found in North Carolina. Jour. Elisha Mitchell Sci. Soc. **23**: 141–149.

———. 1907c. The salamanders of North Carolina. Jour. Elisha Mitchell Sci. Soc. **23**: 150–156.

———. 1907d. A key to the species of frogs and toads liable to occur in North Carolina. Jour. Elisha Mitchell Sci. Soc. **23**: 157–160.

———. 1909. Some notes on the zoology of Lake Ellis, Craven County, North Carolina, with special reference to herpetology. Proc. Biol. Soc. Wash. **22**: 129–138.

———. 1910. Records of some reptiles and batrachians from the southeastern United States. Proc. Biol. Soc. Wash. **23**: 9–18.

———. 1912. Notes on the salamanders of the North Carolina mountains with descriptions of two new forms. Proc. Biol. Soc. Wash. **25**: 135–140.

———. 1915. List of reptiles and amphibians of North Carolina. Jour. Elisha Mitchell Sci. Soc. **30**: 195–206.

———. 1916. The two Raleigh *Amblystomae* compared. Jour. Elisha Mitchell Sci. Soc. **32**: 45–46.

———. 1917a. Some known changes in the land vertebrate fauna of North Carolina. Jour. Elisha Mitchell Sci. Soc. **32**: 176–183.

———. 1917b. The two forms of red *Spelerpes* occurring at Raleigh, N. C. Proc. Biol. Soc. Wash. **30**: 87–88.

———. 1918a. Brief comparison of the herpetological faunas of North Carolina and Virginia. Jour. Elisha Mitchell Sci. Soc. **34**: 146–147.

———. 1918b. Eliminations from and additions to the North Carolina list of reptiles and amphibians. Jour. Elisha Mitchell Sci. Soc. **34**: 148–149.

———. 1918c. *Pituophis melanoleucus* in North Carolina. Copeia **63**: 92.

———. 1920a. The turtles of North Carolina; with a key to the turtles of the eastern United States. Jour. Elisha Mitchell Sci. Soc. **36**: 62–71.

———. 1920b. Notes on *Amphiuma* and *Necturus*. Copeia **77**: 5–7.

———. 1920c. Reproduction of the marbled salamander. Copeia **80**: 25.

———. 1920d. Notes on *Pseudemys scripta* Schoepff, the yellow-bellied terrapin. Copeia **87**: 93–94.

———. 1920e. Notes on *Lampropeltis elapsoides virginiana* Blanchard. Copeia **89**: 106–109.

———. 1921a. Breeding dates of *Ambystoma maculatum* at Raleigh, N. C. Copeia **93**: 26–27.

———. 1921b. The life history of the American newt. Copeia **94**: 31–32.

———. 1922a. Herpetological notes from North Carolina. Copeia **107**: 47–48.

———. 1922b. Herpetological notes from North Carolina (II). Copeia **109**: 63–64.

———. 1923a. North Carolina herpetology. Copeia **114**: 3–4.

———. 1923b. The dwarf salamander at Raleigh, N. C. Copeia **120**: 81–83.

———. 1923c. The copperhead moccasin at Raleigh, N. C. Copeia **124**: 113–116.

———. 1924. The water dogs (*Necturus*) of North Carolina. Jour. Elisha Mitchell Sci. Soc. **40**: 166–168.

———. 1925. The seasonal catch of snakes at Raleigh, N. C. Jour. Elisha Mitchell Sci. Soc. **41**: 100–103.

———. 1926. Revised key and list of the amphibians and reptiles of North Carolina. Jour. Elisha Mitchell Sci. Soc. **42**: 75–93.

———. 1927a. Some records of amphibians and reptiles from North Carolina. Copeia **162**: 10–12.

———. 1927b. Notes on water snakes at Raleigh, N. C. Copeia **162**: 14–15.

———. 1928a. Yellow-cheeked *Desmognathus* from Macon County, N. C. Copeia **166**: 21–23.

———. 1928b. Two new terrapins of the genus *Pseudemys* from the southeastern United States. Jour. Elisha Mitchell Sci. Soc. **44**: 66–69.

———. 1939a. The Amphibians of North Carolina. N. C. Dept. of Agric., Raleigh. (Mimeographed.)

———. 1939b. The Reptiles of North Carolina. N. C. Dept. of Agric., Raleigh. (Mimeographed.)

———. 1943. Amphibians and Reptiles of North Carolina. Carolina Tips, 1939–1943. Carolina Biol. Supply Co.: Elon College, N. C.

BRIMLEY, C. S., AND W. B. MABEE. 1925. Reptiles, amphibians, and fishes collected in eastern North Carolina in the autumn of 1923. Copeia **139**: 14–16.

BRIMLEY, C. S., AND FRANKLIN SHERMAN. 1908. Notes on the life-zones in North Carolina. Jour. Elisha Mitchell Sci. Soc. **24**: 14–22.

BROWN, WALTER C., AND SHERMAN C. BISHOP. 1947. A new species of *Desmognathus* from North Carolina. Copeia **1947**: 163–166.

BURT, CHARLES E. 1931. A Study of the Teiid Lizards of the Genus *Cnemidophorus* with Special Reference to their Phylogenetic Relationships. Smithsonian Inst. Bull. No. 154.

———. 1937. The lizards of the southeastern United States. Trans. Kans. Acad. Sci. **40**: 349–366.

———. 1938. The frogs and toads of the southeastern United States. Trans. Kans. Acad. Sci. **41**: 331–367.

CARR, A. F., JR. 1935. The identity and status of two turtles of the genus *Pseudemys*. Copeia **1935**: 147–148.

———. 1937. The status of *Pseudemys scripta* and *Pseudemys troostii*. Herpetologica **1**: 74–77.

———. 1938. A new subspecies of *Pseudemys floridana*, with notes on the *floridana* complex. Copeia **1938**: 105–109.

———. 1946. Status of the mangrove terrapin. Copeia **1946**: 170–172.

———. 1952. Handbook of Turtles. Comstock Publ. Assoc.: Ithaca, N. Y.

CHADWICK, CLAUDE S. 1940. Some notes on the burrows of *Plethodon metcalfi*. Copeia **1940**: 50.

———. 1944. Observations on the life cycle of the common newt in western North Carolina. Am. Midl. Nat. **32**: 491–494.

CHAMBERLAIN, E. B. 1939. Frogs and toads of South Carolina. Charleston Mus. Leaflet No. 12.

CLIBURN, J. W. 1958. Some southern races of the common water snake, *Natrix sipedon*. Herpetologica **13**: 193–202.

COLES, RUSSELL J. 1915. Alligators in winter. Copeia **17**: 4.

CONANT, ROGER. 1943. The milk snakes of the Atlantic coastal plain. Proc. New England Zool. Club **22**: 3–24.

———. 1949. Two new races of *Natrix erythrogaster*. Copeia **1949**: 1–15.

———. 1956. Common names for North American amphibians and reptiles. The report of the Committee on Herpetological Common Names. Copeia **1956**: 172–185.

———. 1958. A Field Guide to Reptiles and Amphibians. Houghton Mifflin Co.: Boston.

COPE, E. D. 1889. The Batrachia of North America. Bull. U. S. Nat. Mus. No. 34.

———. 1900. The crocodilians, lizards, and snakes of North America. Annual Report Smithsonian Inst., 1889, Pt. II: 153–1294.

COUES, ELLIOT. 1871. Notes on the natural history of Fort Macon, N. C., and vicinity. Proc. Acad. Nat. Sci. Phila. **1871**: 12–49.

COUES, ELLIOT, AND H. C. YARROW. 1879. Notes on the natural history of Fort Macon, N. C., and vicinity. Proc. Acad. Nat. Sci. Phila. **1878**: 21–28.

DAVIS, HARRY T., AND C. S. BRIMLEY. 1942. Poisonous-Snakes of the Eastern United States with First-Aid Guide. N. C. State Museum, Raleigh.

DEPOE, CHARLES E., AND JOHN B. FUNDERBURG. 1959. The river frog, *Rana hecksheri* Wright, in North Carolina. Jour. Elisha Mitchell Sci. Soc. **75**: 11–12.

DICKERSON, MARY C. 1913. The Frog Book. Doubleday, Page and Co.: Garden City, N. Y.

DITMARS, RAYMOND L. 1908. The Reptile Book. Doubleday, Page and Co.: Garden City, N. Y.

DOWLING, HERNDON G. 1950. Studies of the Black Swamp Snake, *Seminatrix pygaea* (Cope), with Descriptions of Two New Subspecies. Univ. Mich. Misc. Publ. Mus. Zool. No. 76.

———. 1952. A Taxonomic Study of the Ratsnakes, Genus *Elaphe* Fitzinger. IV. A Check List of the American Forms. Univ. Mich. Occ. Papers Mus. Zool. No. 541.

DUNN, EMMETT R. 1917a. The salamanders of the genera *Desmognathus* and *leurognathus*. Proc. U. S. Nat. Mus. **53**: 393–433.

———. 1917b. Reptile and amphibian collections from the North Carolina mountains, with especial reference to salamanders. Am. Mus. Nat. Hist. **37** (23): 593–634.

———. 1920. Some reptiles and amphibians from Virginia, North Carolina, Tennessee and Alabama. Proc. Biol. Soc. Wash. **33**: 129–138.

———. 1926. The Salamanders of the Family Plethodontidae. Smith College Anniv. Publ., Northampton, Mass.

———. 1927a. A new mountain race of *Desmognathus*. Copeia **164**: 84–86.

———. 1927b. On the relationships of certain plethodont salamanders. Copeia **165**: 102–106.

———. 1940. The races of *Ambystoma tigrinum*. Copeia **1940**: 154–162.

EATON, THEODORE H., JR. 1953. Salamanders of Pitt County, North Carolina. Jour. Elisha Mitchell Sci. Soc. **69**: 49–53.

———. 1954. *Desmognathus perlapsus* Neill in North Carolina. Herpetologica **10**: 41–43.

———. 1956. Larvae of some Appalachian plethodontid salamanders. Herpetologica **12**: 303–311.

EATON, THEODORE H., JR., AND G. T. EATON. 1956. A new locality for the green salamander and wood frog in North Carolina. Herpetologica **12**: 312.

ENGELS, WILLIAM L. 1942. Vertebrate fauna of North Carolina coastal islands. Am. Midl. Nat. **28**: 273–304.

———. 1949. The blue-tailed skinks (*Eumeces*) of two North Carolina coastal islands. Copeia **1949**: 269–271.

———. 1952. Vertebrate fauna of North Carolina coastal islands. Am. Midl. Nat. **47**: 702–742.

FAHY, WILLIAM E. 1954. Loggerhead turtles, *Caretta caretta caretta*, from North Carolina. Copeia **1954**: 157–158.

FUNDERBURG, JOHN B. 1953. The Georgia tree frog, *Hyla gratiosa* Leconte, in North Carolina. Herpetologica **9**: 176.

———. 1955. The amphibians of New Hanover County, North Carolina. Jour. Elisha Mitchell Sci. Soc. **71**: 20–28.

———. 1959. The yellow-lipped snake, *Rhadinaea flavilata* Cope, in North Carolina. Jour. Elisha Mitchell Sci. Soc. **74**: 135–136.

GLOYD, HOWARD K. 1935a. The Subspecies of *Sistrurus miliarius*. Univ. Mich. Occ. Papers Mus. Zool. No. 322.

———. 1935b. The cane-brake rattlesnake. Copeia **1935**: 175–178.

———. 1940. The Rattesnakes, Genera *Sistrurus* and *Crotalus*. Chicago Acad. Sci., Spec. Publ. No. 4.

GLOYD, HOWARD K., AND ROGER CONANT. 1943. A synopsis of the American forms of *Agkistrodon* (copperheads and moccasins). Bull. Chicago Acad. Sci. **7**: 147–170.

GORDON, ROBERT E., AND RICHARD L. SMITH. 1949. Notes on the life history of the salamander *Aneides aeneus*. Copeia **1949**: 173–175.

GOSNER, KENNETH L., AND IRVING H. BLACK. 1956. Notes on amphibians from the upper coastal plain of North Carolina. Jour. Elisha Mitchell Sci. Soc. **72**: 40–47.

——— AND ———. 1957. Notes on the life

history of Brimley's chorus frog. Herpetologica **13**: 249–254.

GRAY, I. E. 1941. Amphibians and reptiles of the Duke Forest and vicinity. Am. Midl. Nat. **25**: 652–658.

GREEN, N. BAYARD, AND NEILL D. RICHMOND. 1944. Courtship of *Plethodon metcalfi*. Copeia **1944**: 256.

GROBMAN, ARNOLD B. 1941a. A Contribution to the Knowledge of Variation in *Opheodrys vernalis* (Harlan), with the Description of a New Subspecies. Univ. Mich. Misc. Publ. Mus. Zool. No. 50.

———. 1941b. Variation of the salamander *Pseudotriton ruber*. Copeia **1941**: 179.

———. 1944. The distribution of the salamanders of the genus *Plethodon* in eastern United States and Canada. Ann. N. Y. Acad. Sci. **45**: 261–316.

———. 1950. The Distribution of the Races of *Desmognathus fuscus* in the southern states. Nat. Hist. Miscellanea **70**: 1–8.

HAIRSTON, NELSON G. 1947. The Gyrinophilus Salamanders of Mt. Mitchell, North Carolina. Nat. Hist. Miscellanea, No. 7.

———. 1949. The local distribution and ecology of the plethodontid salamanders of the southern Appalachians. Ecol. Monographs **19**: 47–73.

———. 1950. Intergradation in Appalachian salamanders of the genus *Plethodon*. Copeia **1950**: 262–273.

HARDY, JERRY D., JR. 1952. The crowned snake, *Tantilla coronata coronata*, in North Carolina. Copeia **1952**: 188.

HARPER, FRANCIS. 1935. Records of some amphibians in the southeastern states. Am. Midl. Nat. **16**: 275–310.

———. 1937. A season with Holbrook's chorus frog (*Pseudacris ornata*). Am. Midl. Nat. **18**: 260–272.

HIGHTON, RICHARD, AND ARNOLD B. GROBMAN. 1956. Two new salamanders of the genus *Plethodon* from the southeastern United States. Herpetologica **12**: 185–188.

HOFFMAN, RICHARD L. 1945. Range extension for *Eumeces inexpectatus* Taylor. Proc. Biol. Soc. Wash. **58**: 131–132.

———. 1951. A new subspecies of salamander from Virginia. Jour. Elisha Mitchell Sci. Soc. **67**: 249–254.

———. 1957a. A new subspecies of the teiid lizard *Cnemidophorus sexlineatus* (Linnaeus) from eastern United States. Jour. Wash. Acad. Sci. **47**: 153–156.

———. 1957b. A new name for the race-runner lizard of the Middle Atlantic States (Teiidae). Jour. Wash. Acad. Sci. **47**: 423.

HOFFMAN, RICHARD L., AND LESLIE HUBRICHT. 1954. Distributional records for two species of *Plethodon* in the southern Appalachians. Herpetologica **10**: 191–193.

HOWELL, THELMA. 1950. Red blood cell size in the male frog, *Rana catesbeiana* Shaw. Jour. Tenn. Acad. Sci. **25**: 237–241.

HOWELL, THELMA, AND ANN HAWKINS. 1954. Variation in topotypes of the salamander *Plethodon jordani melaventris*. Copeia **1954**: 32–36.

HUHEEY, JAMES E. 1959. Distribution and variation in the glossy water snake, *Natrix rigida* (Say). Copeia **1959**: 303–311.

HUMPHRIES, ROBERT L. 1951. The woodfrog in Georgia. Copeia **1951**: 183.

HUTCHINSON, VICTOR H. 1956. Notes on the Plethodontid Salamanders, *Eurycea lucifuga* (Rafinesque) and *Eurycea longicauda longicauda* (Green). Occ. Papers Nat. Spelological Soc. No. 3.

JANSON, HARVEY S. 1953. Skittering locomotion in the frog *Hyla cinerea cinerea*. Copeia **1953**: 62.

JOHNSON, RICHARD M. 1954. The painted turtle, *Chrysemys picta picta*, in eastern Tennessee. Copeia **1954**: 298–299.

KAUFFELD, CLARK F. 1937. The status of the leopard frogs, *Rana brachycephala* and *Rana pipiens*. Herpetologica **1**: 84–87.

KELLOGG, REMINGTON. 1929. The Habits and Economic Importance of Alligators. U.S.D.A. Tech. Bull. No. 147. Washington, D. C.

KING, WILLIS. 1936. A new salamander (*Desmognathus*) from the southern Appalachians. Herpetologica **1**: 57–60.

———. 1939. A survey of the herpetology of Great Smoky Mountains National Park. Am. Midl. Nat. **21**: 531–582.

———. 1944. Additions to the list of amphibians and reptiles of Great Smoky Mountains National Park. Copeia **1944**: 255.

KLAUBER, LAURENCE M. 1936. A key to the rattlesnakes with summary of characteristics. Trans. San Diego Soc. Nat. Hist. **8**: 185–276.

LEWIS, THOMAS H. 1946. Reptiles and amphibians of Smith Island, N. C. Am. Midl. Nat. **36**: 682–684.

MCCONKEY, EDWIN H. 1954. A systematic study of the North American lizards of the genus *Ophisaurus*. Am. Midl. Nat. **51**: 133–171.

MALNATE, EDMOND. 1939. A study of the yellow-lipped snake, *Rhadinaea flavilata* (Cope). Zoologica **24**: 359–367.

MARTOF, BERNARD. 1955a. Observations on the life history and ecology of the amphibians of the Athens area, Georgia. Copeia **1955**: 166–170.

———. 1955b. Some records of reptiles of Georgia. Copeia **1955**: 302–305.

———. 1956. Three New Subspecies of *Leurognathus marmorata* from the Southern Appalachian Mountains. Univ. Mich. Occ. Papers Mus. Zool. No. 575.

MARTOF, BERNARD, AND ROBERT L. HUMPHRIES. 1955. Observations on some amphibians from Georgia. Copeia **1955**: 245–248.

——— AND ———. 1959. Geographic variation in the wood frog, *Rana sylvatica*. Am. Midl. Nat. **61**: 350–389.

MECHAM, JOHN S. 1954. Geographic variation in

the green frog, *Rana clamitans* Latreille. Texas Jour. Sci. **6**: 1–25.

MITTLEMAN, M. B. 1947. American Caudata. I. Geographic variation in *Manculus quadridigitatus*. Herpetologica **3**: 209–224.

———. 1951. American Caudata. VII. Two new salamanders of the genus *Plethodon*. Herpetologica **7**: 105–112.

MITTLEMAN, M. B., AND HARRY G. JOPSON. 1941. A New Salamander of the Genus *Gyrinophilus* from the Southern Appalachians. Smithsonian Misc. Coll. 101.

MOORE, JOHN A. 1942. The role of temperature in speciation in frogs. Biological Symposia **6**: 189–213.

MUCHMORE, WILLIAM B. 1955. Brassy flecking in the salamander *Plethodon c. cinereus*, and the validity of *Plethodon huldae*. Copeia **1955**: 170–172.

MYERS, GEORGE S. 1924. Amphibians and reptiles from Wilmington, N. C. Copeia **131**: 59–62.

NEILL, WILFRED T. 1947. Size and habits of the cottonmouth moccasin. Herpetologica **3**: 203–205.

———. 1949a. Forms of *Ophisaurus* in the southeastern United States. Herpetologica **5**: 97–100.

———. 1949b. A new subspecies of rat snake (genus *Elaphe*), and notes on related forms. Herpetologica **5**: 1–12.

———. 1950. Taxonomy, nomenclature, and distribution of southeastern cricket frogs, genus *Acris*. Am. Midl. Nat. **43**: 152–156.

———. 1951. The taxonomy of North American soft-shelled turtles, genus *Amyda*. Ross Allen's Reptile Institute, **1**: 7–24.

———. 1954. Ranges and taxonomic allocations of amphibians and reptiles in the southeastern United States. Ross Allen's Reptile Institute, **1**: 75–96.

NOBLE, G. KINGSLEY. 1927. The Plethodontid Salamanders; Some Aspects of their Evolution. Am. Mus. Novitates No. 249.

NOBLE, G. KINGSLEY, AND RUTH C. NOBLE. 1923. The Anderson tree frog (*Hyla andersonii* Baird). Zoologica **2**: 414–455.

PALMER, LAURENCE. 1939. Some common reptiles. Nature Magazine. April 1939.

PALMER, W. M. 1959. A second record of the glossy water snake in North Carolina. Herpetologica **15**: 47.

PERKINS, C. B. 1949. A Key to the Snakes of the United States. Bull. Zool. Soc. San Diego No. 24.

PICKENS, ANDREW L. 1927. Intermediates between *Bufo fowleri* and *B. americanus*. Copeia **162**: 25–26.

POPE, CLIFFORD H. 1924. Notes on North Carolina Salamanders with Especial Reference to the Egg-laying Habits of *Leurognathus* and *Desmognathus*. Am. Mus. Novitates No. 153.

———. 1928. Some Plethodontid Salamanders from North Carolina and Kentucky with the Description of a New Race of *Leurognathus*. Am. Mus. Novitates No. 306.

———. 1939. Turtles of the United States and Canada. Alfred A. Knopf: New York, N. Y.

———. 1949. The Salamander *Desmognathus quadramaculatus amphileucus* Reduced to Synonomy. Nat. Hist. Miscellanea No. 44.

POPE, CLIFFORD H., AND NELSON G. HAIRSTON. 1947. The distribution of *Leurognathus*, a southern Appalachian genus of salamander. Fieldiana Zool., Chicago Nat. Hist. Mus. **31**: 155–162.

——— AND ———. 1948. Two new subspecies of the salamander, *Plethodon shermani*. Copeia **1948**: 106–107.

RABB, GEORGE B. 1955. Observations on the identity of the salamander *Plethodon huldae*. Copeia **1955**: 261–262.

RANKIN, JOHN S. 1937. An ecological study of parasites of some North Carolina salamanders. Ecol. Monographs **7**: 169–269.

REESE, ROBERT W. 1949. The Occurrence of *Eumeces inexpectatus* in Kentucky. Nat. Hist. Miscellanea No. 39.

———. 1950. The Identity of the Salamander *Gyrinophilus danielsi*, with Description of a New Subspecies. Nat. Hist. Miscellanea No. 63.

ROBERTSON, WILLIAM B., AND EDWARD L. TYSON. 1950. Herpetological notes from eastern North Carolina. Jour. Elisha Mitchell Sci. Soc. **66**: 130–147.

SCHMIDT, KARL PATTERSON. 1916. Notes on the herpetology of North Carolina. Jour. Elisha Mitchell Sci. Soc. **32**: 33–37.

———. 1953. A Check List of North American Amphibians and Reptiles. 6th ed. University of Chicago Press: Chicago, Ill.

SCHMIDT, KARL PATTERSON, AND D. DWIGHT DAVIS. 1941. Fieldbook of Snakes of the United States and Canada. G. P. Putnam's Sons: New York.

SCHWARTZ, ALBERT. 1953. A new subspecies of crowned snake (*Tantilla coronata*) from the southern Appalachian mountains. Herpetologica **9**: 153–157.

———. 1954. The salamander *Aneides aeneus* in South Carolina. Copeia **1954**: 296–298.

———. 1955. The chorus frog *Pseudacris brachyphona* in North Carolina. Copeia **1955**: 138.

———. 1956. The Relationships and Nomenclature of the Soft-shelled Turtles (genus *Trionyx*) of the Southeastern United States. Charleston Mus. Leaflet No. 26.

SCHWARTZ, ALBERT, AND RICHARD ETHERIDGE. 1954. New and additional herpetological records from the North Carolina coastal plain. Herpetologica **10**: 167–171.

SCHWARTZ, ALBERT, AND JULIAN HARRISON, III. 1956. A new subspecies of gopher frog, (*Rana capito* Leconte). Proc. Biol. Soc. Wash. **69**: 135–144.

SIMMONS, ROBERT, AND JERRY D. HARDY, JR. 1959. The river-swamp frog, *Rana hecksheri* Wright, in North Carolina. Herpetologica **15**: 36–37.

SMITH, HOBART M. 1938a. A review of the snake genus *Farancia*. Copeia **1938**: 110–117.

———. 1938b. Remarks on the Status of the

Subspecies of *Sceloporus undulatus*, with Descriptions of New Species and Subspecies of the *undulatus* Group. Univ. Mich. Occ. Papers Mus. Zool. No. 387.

———. 1946a. The systematic status of *Eumeces pluvialis* Cope, and noteworthy records of other amphibians and reptiles from Kansas and Oklahoma. Univ. Kansas Mus. Nat. Hist. **1**: 85–89.

———. 1946b. Handbook of Lizards. Comstock Publ. Co.: Ithaca, N. Y.

SMITH, PHILIP W., AND HOBART M. SMITH. 1952. Geographic variation in the lizard *Eumeces anthracinus*. Univ. Kansas Sci. Bull. **34**: 679–694.

SNYDER, RICHARD C. 1946. *Plethodon welleri* from Flat Top Mountain, North Carolina. Copeia **1946**: 174.

STEJNEGER, LEONHARD. 1903. Rediscovery of one of Holbrook's salamanders. Proc. U. S. Nat. Mus. **28**: 557–558.

———. 1906. A new salamander from North Carolina. Proc. U. S. Nat. Mus. **30**: 559–562.

———. 1944. Notes on the American soft-shelled turtles with special reference to *Amyda agassizi*. Harvard Univ. Bull. Mus. Comp. Zool. **94**: 1–75.

STEJNEGER, LEONHARD, AND THOMAS BARBOUR. 1923. A Check List of North American Amphibians and Reptiles. 2d. ed. Harvard University Press: Cambridge, Mass.

——— AND ———. 1933. A Check List of North American Amphibians and Reptiles. 3d. ed. Harvard University Press: Cambridge, Mass.

——— AND ———. 1939. A Check List of North American Amphibians and Reptiles, 4th ed. Harvard University Press: Cambridge, Mass.

STEWART, MARGARET MCBRIDE. 1956. The separate effects of food and temperature differences on development of marbled salamander larvae. Jour. Elisha Mitchell Sci. Soc. **72**: 47–56.

STULL, OLIVE G. 1932. An Annotated List of the Forms of the Genus *Pituophis*. Univ. Mich. Occ. Papers Mus. Zool. No. 250.

———. 1940. Variations and Relationships in the Snakes of the Genus *Pituophis*. Bull. U. S. Nat. Mus. No. 175.

TAYLOR, EDWARD H. 1932. *Eumeces inexpectatus*: A new American lizard of the family Scincidae. Univ. Kansas Sci. Bull. **20**: 251–261.

———. 1935. A taxonomic study of the cosmopolitan Scincid lizards of the genus *Eumeces* with an account of the distribution and relationships of its species. Univ. Kansas Sci. Bull. **23**: 1–643.

———. 1943. Mexican lizards of the genus *Eumeces*, with comments on the recent literature on the genus. Univ. Kansas Sci. Bull. **29**: 269–300.

TRAPIDO, HAROLD. 1944. The snakes of the genus *Storeria*. Am. Midl. Nat. **31**: 1–84.

THURROW, GORDON R. 1956. A new subspecies of *Plethodon welleri*, with notes on other members of the genus. Am. Midl. Nat. **55**: 343–356.

VIOSCA, PERCY, JR. 1924. A contribution to our knowledge of the water snakes. Copeia **126**: 3–13.

———. 1937. A tentative revision of the genus *Necturus* with descriptions of three new species from the southern gulf drainage area. Copeia **1937**: 120–138.

VOLPE, E. PETER. 1952. Physiological evidence for natural hybridization of *Bufo americanus* and *Bufo fowleri*. Evolution **6**: 395–406.

———. 1955. Intensity of reproductive isolation between sympatric and allopatric populations of *Bufo americanus* and *Bufo fowleri*. Am. Midl. Nat. **89**: 303–318.

WALKER, CHARLES F. 1931. Description of a new salamander from North Carolina. Proc. Junior Soc. Nat. Sci. Cincinnati **2**: 48–51.

WELLER, W. H. 1931. A preliminary list of the salamanders of the Great Smoky Mts. of North Carolina and Tennessee. Proc. Junior Soc. Nat. Sci. Cincinnati **2**: 21–32.

WERLER, JOHN E., AND JAMES MCCALLION. 1951. Notes on a collection of reptiles and amphibians from Princess Anne County, Virginia. Am. Midl. Nat. **45**: 245–252.

WOOD, JOHN THORNTON. 1946a. *Graptemys geographica* (LeSueur) added to the herpetofaunal list of Great Smoky Mountains National Park. Copeia **1946**: 168.

———. 1946b. Measurements of a giant *Pseudotriton montanus montanus* larva from Great Smoky Mountains National Park. Copeia **1946**: 168.

———. 1947. Notes on North Carolina salamanders. Copeia **1947**: 273–274.

———. 1949. *Eurycea bislineata wilderae* Dunn. Herpetologica **5**: 61–62.

WRIGHT, ALBERT HAZEN. 1918. Notes on *Clemmys*. Proc. Biol. Soc. Wash. **31**: 51–58.

———. 1932. Life-histories of the Frogs of Okefenokee Swamp, Georgia. The Macmillan Co.: New York.

WRIGHT, ALBERT HAZEN, AND ANNA ALLEN WRIGHT. 1949. Handbook of Frogs and Toads of the United States and Canada. Comstock Publ. Co.: Ithaca, New York.

——— AND ———. 1957. Handbook of Snakes of the United States and Canada. 2 vols. Comstock Publ. Co.: Ithaca, New York.

YARROW, H. C. 1882. Check List of North American Reptilia and Batrachia, with Catalogue of Specimens in the U. S. National Museum. Bull. U. S. Nat. Mus. No. 24. Washington, D. C.

OCCURRENCE OF A LEUCOANTHOCYANIN IN HYPOCOTYLS OF ETIOLATED BUCKWHEAT SEEDLINGS[1]

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Many plants have been reported to contain colorless substances which give rise to anthocyanidins upon treatment with hot or concentrated acids. From a study in which plant extracts were simply heated with acid and the resulting red-colored materials examined without purification or isolation, Rosenheim (1920) termed the colorless precursors "leucoanthocyanins." Suggestions concerning the chemical structure of leucoanthocyanins were made by Robinson and Robinson (1933, 1935), but the first clear characterization of one of these substances as a flavan-3, 4-diol was accomplished by King and co-workers (1953, 1955). The Robinsons (1933, 1937) decided that the red products were indeed anthocyanidins largely on the basis of their solubilities and color reactions, although in one case cyanidin was actually isolated. In recent years paper-chromatographic separations and determinations of absorption spectra have been used in establishing this conclusion firmly (Simmonds, 1954; Bate-Smith, 1954).

Mature plants of buckwheat have been shown to contain leucoanthocyanins in several organs. By chromatographic and spectral analysis Bate-Smith (1954) has demonstrated in the roots of this plant a substance which yields cyanidin upon acid treatment. On the basis of a nonspecific color test Bate-Smith and Lerner (1954) have reported that the green leaves contain leucoanthocyanin material. Whether leucoanthocyanins are present in buckwheat seedlings which have developed in the dark, however, has been a matter of some debate. From the appearance of a red color in acid-treated extracts Jonesco (1930) and Kuilman (1930) inferred that leucoanthocyanins occur in etiolated seedlings. After a study of solubilities and color reactions Karstens (1939) decided that the red product was not an anthocyanidin and that therefore leucoanthocyanins are not present in the etiolated seedlings.

Since the hypocotyls of etiolated buckwheat seedlings are a convenient material to use in studies of anthocyanin formation, and since the biosynthetic relations between anthocyanins and leucoanthocyanins are still uncertain (Bogorad, 1958), decision concerning the occurrence of the leuco-substances in these plants is of interest. This report describes a study designed to provide more information concerning this problem.

Materials and Methods

Achenes of *Fagopyrum sagittatum* Gilib. (*F. esculentum* Moench), Japanese variety, were soaked in distilled water for one hour, then spread on four layers of wet Whatman No. 1 filter paper. Each quadruple filter-paper sheet was supported atop two inverted halves of 10-cm.-square plastic petri dishes which floated on distilled water in a Pyrex dish 17 × 25 × 5 cm. Sufficient water was added initially to the dish to insure an adequate moisture supply during the entire growth period. Germination and growth occurred in darkness at 25°C. in a constant-temperature cabinet.

In the preparation of extracts only the hypocotyls of the plants were used. Etiolated seedlings were taken for extraction directly from the dark cabinet at an age of five days. No anthocyanin is formed in buckwheat seedlings unless they are exposed to light. Accordingly, for the preparation of extracts of anthocyanin-containing seedlings, five-day-old plants were removed from the growth cabinet, exposed for four hours to light from two 40-watt daylight fluorescent lamps held at a distance of 15 cm., and then replaced in the dark cabinet for 24 hours. At the end of this time large quantities of anthocyanin pigment were present in the hypocotyls. The use of plants from three to ten days old led to results which were not qualitatively different from those obtained with five-day-old plants. Extracts were made with either methanol or water, each containing an appropriate concentration of HCl. The hypocotyl material was cut transversely into small segments, placed in test tubes containing the extraction solvent, and crushed with a glass rod.

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Extracts used in paper chromatography were prepared in aqueous 0.3N HCl. In order to bring about rapid conversion of leucoanthocyanin to anthocyanidin, the test tubes containing the extracts were held for 20 minutes in a bath of boiling water, cooled, filtered, and shaken with sufficient isoamyl alcohol to give a thin supernatant layer which contained the anthocyanidin (Bate-Smith, 1954). Extracts of *Rosa* petals were used as sources of cyanidin for chromatographic comparisons; these were prepared in the same way as the buckwheat extracts. The isoamyl alcohol was then spotted on strips of Whatman No. 3 mm. filter paper 40 cm. long. One-dimensional descending chromatography was carried out after equilibration of the paper strips with the solvent vapor for 24 hours. The solvents used were formic acid–2N HCl (1-1 vol./vol.), acetic acid–conc. HCl–water (30–3–10, vol./vol./vol.), and the eight systems previously employed in a study of buckwheat pigments (Troyer, 1958). Anthocyanidin spots were located by examining the completed chromatograms in visible and ultraviolet lights.

Extracts used in the measurement of absorption spectra were prepared in 0.3N HCl in methanol. These were heated in boiling water for 20 minutes, cooled, filtered, and used directly. Light-absorption measurements were made with a Bausch and Lomb Spectronic 20 colorimeter.

Extracts used in studying the rate of color development in different concentrations of HCl were prepared in methanol-HCl from both etiolated and anthocyanin-containing hypocotyls. All samples contained the same number of hypocotyls, and five replicates were used for each HCl concentration. The test tubes containing the solvent and the crushed hypocotyl material were placed at 13°C. for extraction. After 24 hours the extracts were filtered, transferred to colorimeter tubes calibrated to contain a known volume, and held in the dark at either 13° or 25°C. From time to time the samples were made to volume by addition of extraction solvent as necessary, and absorbance values measured at 535 mμ, an absorption maximum of the buckwheat anthocyanidin. Less frequently complete absorption spectra were determined in the range 350 to 650 mμ.

Results

In preliminary studies the experience of Karstens (1939) with buckwheat extracts was confirmed. Heating the acidic yellow extract of etiolated seedlings always resulted in the formation of a dark red anthocyanin-like color in the solution. However, the solubilities of the bulk of this colored material in various solvents were in certain respects different from those of anthocyanidins. Also, the color reactions shown by the crude red extracts were not what one would expect; in particular, the color with sodium carbonate was gray instead of the blue which develops in anthocyanidin solutions. It was on the basis of these properties of the crude extracts that Karstens concluded that no leucoanthocyanin is present in the etiolated seedlings.

However, application of paper chromatography, a technique not available to Karstens, showed that a substance is present in the heat-treated acidic extracts of etiolated hypocotyls which behaves chromatographically like the anthocyanidin of anthocyanin-containing hypocotyls. No R_f values are reported here because considerable variation in these quantities was observed in different series of chromatograms. The critical result is that in a given series the two materials showed R_f values which were almost identical, never differing from each other by more than 0.02 R_f unit. This was true for every solvent used. Also, both materials moved in all solvents like the cyanidin contained in extracts of *Rosa* petals. No such anthocyanidin material was present in the extracts of etiolated hypocotyls unless these were heated or allowed to stand at 25°C. for some time.

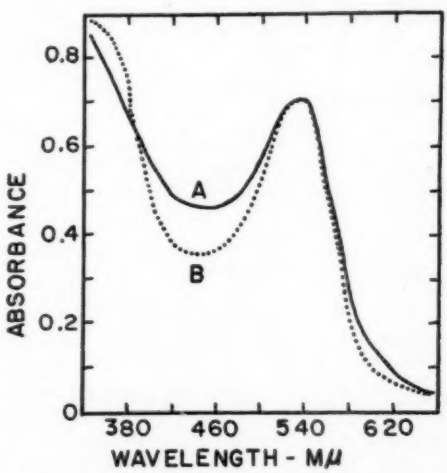

FIG. 1. Absorption spectra of heated crude extracts of etiolated (A) and anthocyanin-containing (B) buckwheat hypocotyls. The solvent was 0.3N HCl in methanol. For purposes of comparison the two curves are made to coincide in absorbance at 535 mμ.

Absorption spectra of crude, heat-treated, acidic extracts of etiolated and anthocyanin-containing hypocotyls are similar in the range 350 to 650 mμ (Fig. 1). Both kinds of extract show a single absorption maximum at 535 mμ and a region of minimum absorption near 440 mμ. Repeated measurements of spectra of such crude extracts never yielded evidence of a second absorption maximum near 459 mμ, such as was observed by Raudnitz (1958) and shown by him to be due to the presence of a humic acid derivative formed during heating with acid.

One study was designed to show the importance of temperature and HCl concentration on the reaction by which anthocyanidin is formed from leucoanthocyanin in extracts of etiolated hypocotyls. In this case extracts held at 13°C. showed no trace of red color over a period of 261 hours, regardless of the concentration of HCl used (0.3N, 1.6N, or 3.3N). The absorbances of the extracts at 535 mμ remained at the initial levels, and no anthocyanidin spot could be found on paper chromatograms. Extracts of etiolated hypocotyls held at 25°C., however, gradually became red in color, a visible change measured quantitatively by increased absorbance at 535 mμ. As the curves in Figure 2 show, both the initial rate of increase in absorbance of these extracts and the final maximum reached were greater at higher levels of HCl. After 48 hours all extracts showed absorption spectra similar to those of Figure 1.

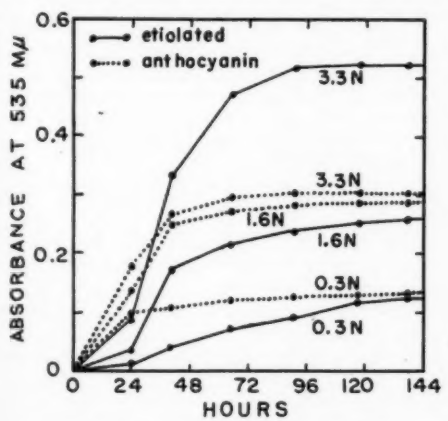

FIG. 2. Rate of development of color at 25°C. in crude extracts of etiolated and anthocyanin-containing buckwheat hypocotyls and the effect of HCl normality in the methanol extraction solvent. Each point represents the mean of five samples.

The behavior of extracts of anthocyanin-containing hypocotyls was similar, although these extracts initially contained several anthocyanins and the anthocyanidin (Troyer, 1958). Extracts kept at 13°C. showed no change in light absorption or chromatographic properties. Those held at 25°C. increased in absorbance at 535 mμ with time in a manner dependent upon the HCl concentration (Fig. 2). In this case, however, there was little difference in the results obtained with 1.6N and 3.3N HCl. Paper chromatograms of the extracts showed a simultaneous increase in the quantity of anthocyanidin and decrease in the quantities of anthocyanins.

Discussion

From the results of the chromatographic studies and light-absorption measurements described here, it may reasonably be concluded that the hypocotyls of etiolated buckwheat seedlings contain a leucoanthocyanin. This substance is present in acidic extracts of such hypocotyls and under suitable temperature conditions apparently is changed to cyanidin, the anthocyanidin characteristic of the buckwheat plant. In the author's opinion, the failure of Karstens (1939) to detect this result probably results from the fact that he was dependent upon solubility and color-reaction techniques. The amount of anthocyanidin formed in the extracts is small, and the solubilities and color reactions observed by Karstens and by the author in crude extracts were probably greatly affected by the presence of other materials. The possibility exists that one such material might be the kind of red pigment found by Raudnitz (1958) in heated acidic plant extracts; this substance was reported to show two light-absorption maxima at 548 and 459 mμ. Two such peaks were never observed by the author in buckwheat extracts; however, the first might have been masked by the anthocyanidin maximum at 535 mμ, and the second might be responsible for the fact that extracts of etiolated hypocotyls show greater absorption in the range 420 to 480 mμ than do extracts of anthocyanin-containing hypocotyls (Fig. 1).

The study on the rate of color development shows that even at room temperature the leucoanthocyanin-anthocyanidin conversion may occur in time in acidic extracts. Accordingly, such extracts of plant materials should be held at a low temperature if such a change is to be prevented.

Since in the rate study the numbers of hypocotyls used and the volume of extract

employed were the same for all samples, the absorbance values shown in Figure 2 may be taken as rough approximations of the quantities of anthocyanidin present in the various extracts. It is interesting, then, to compare the final absorbance values reached in samples derived from etiolated and anthocyanin-containing hypocotyls. For the 0.3N and 1.6N concentrations of HCl the values for the two kinds of extract were about the same. In 3.3N HCl, however, the value for the etiolated hypocotyls is considerably higher than that for anthocyanin-containing hypocotyls. This result might mean that more anthocyanidin may be produced by leucoanthocyanin conversion in the first case than by anthocyanin hydrolysis in the second.

Since the situation in buckwheat hypocotyls is apparently rather simple—one leucoanthocyanin in the etiolated material which yields, after suitable treatment, the same anthocyanidin usually found in light-treated hypocotyls—this plant offers a very convenient object to use in studying the biosynthetic relations of leucoanthocyanins and anthocyanins. One basic question which would need to be answered in such an investigation is whether any leucoanthocyanin occurs in anthocyanin-containing hypocotyls; if not, the possible conversion of leucoanthocyanin to anthocyanin at some time following exposure to light might be examined. The present study, of course, offers no result by which this question might be answered

Summary

Acidic extracts of hypocotyls of etiolated buckwheat seedlings contain a substance which is converted at suitable temperatures to cyanidin, the anthocyanidin characteristic of this plant. The anthocyanidin product is identifiable by paper chromatography and measurement of absorption spectra. The rate of the leucoanthocyanin-anthocyanidin conversion and the total amount of anthocyanidin formed are influenced by temperature and by the concentration of acid in the extraction solvent.

LITERATURE CITED

BATE-SMITH, E. C. 1954. Leuco-anthocyanins. I. Detection and identification of anthocyanidins formed from leuco-anthocyanins in plant tissues. Biochem. Jour. **58**: 122–125.

BATE-SMITH, E. C., AND N. H. LERNER. 1954. Leuco-anthocyanins. 2. Systematic distribution of leuco-anthocyanins in leaves. Biochem. Jour. **58**: 126–132.

BOGORAD, L. 1958. The biogenesis of flavonoids. Ann. Rev. Plant. Physiol. **9**: 417–448.

JONESCO, S. 1930. Les anthocyanidines et les leucoanthocyanidines chez les végétaux. Ann. Sci. Nat. Sér. 10, Botan. **12**: 249–255.

KARSTENS, W. K. H. 1939. Anthocyanin and anthocyanin formation in seedlings of *Fagopyrum esculentum* Moench. Rec. trav. bot. néerl. **36**: 85–179.

KING, F. E., AND W. BOTTOMLEY. 1953. The isolation from *Acacia melanoxylon* of a flavan-3:4-diol and its possible bearing on the constitution of the phlobatannins. Chem. and Ind. **1953**: 1368.

KING, F. E., AND J. W. CLARK-LEWIS. 1955. The constitution and synthesis of leucoanthocyanidins. Jour. Chem. Soc. **1955**: 3384–3388.

KUILMAN, L. W. 1930. Physiologische Untersuchungen über die Anthocyane. Rec. trav. bot. néerl. **27**: 287–416.

RAUDNITZ, H. 1958. Concerning a pigment commonly attributed to the presence of leucoanthocyanin. Science **128**: 782.

ROBINSON, G. M. 1937. Leucoanthocyanins. III. Formation of cyanidin chloride from a constituent of the gum of *Butea frondosa*. Jour. Chem. Soc. **1937**: 1157–1160.

ROBINSON, G. M., AND R. ROBINSON. 1933. A survey of anthocyanins. III. Notes on the distribution of leuco-anthocyanins. Biochem. Jour. **27**: 206–212.

——— AND ———. 1935. Leuco-anthocyanins and leuco-anthocyanidins. I. The isolation of peltogynol and its molecular structure. Jour. Chem. Soc. **1935**: 744–752.

ROSENHEIM, O. 1920. XXI. Observations on anthocyanins. 1. The anthocyanins of the young leaves of the grape vine. Biochem. Jour. **14**: 178–188.

SIMMONDS, N. W. 1954. Anthocyanins in bananas. Nature **173**: 402–403.

TROYER, J. R. 1958. Anthocyanin pigments of buckwheat hypocotyls. Ohio Jour. Sci. **58**: 187–188.

OPISTHOBRANCHIA FROM NORTH CAROLINA

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During his study of the fauna of oyster beds at the Duke University Marine Laboratory at Beaufort, North Carolina, and on other occasions, Dr. Harry W. Wells of Duke University collected opisthobranchs which he sent to Mrs. Eveline du Bois-Reymond Marcus and me for classification, adding notes on their colors in life (The material has been returned to him). In addition, two samples came from Ocean City, Maryland. The scarcity of reports on opisthobranchs from E. Forbes' Carolinian province (Johnson, 1934: 8) accounts for the high proportion of zoogeographic and systematic novelties.

We are grateful to Dr. Wells for a linguistic revision of the manuscript and for proofreading.

1. *Hermaea dendritica* (Alder and Hancock, 1843)

Occurrence: Beaufort, N. C., 25. III. 1956; two specimens. Further distribution: Mediterranean; eastern Atlantic northward to southern Norway—Bergen; southern and middle Japan.

Odhner's reproduction of Lovén's figures (1907, pl. 3, figs. 28, 29) shows that *H. venosa* is *dendritica*, but *bifida* (Montagu, 1815) is the type species of *Hermaea* (Fischer, 1887: 542). If the absence (*bifida*) or presence (*dendritica*) of an albumen gland in the cerata were considered a generic or subgeneric character, *dendritica* would be removed from *Hermaea*. However Pruvot-Fol's assignment (1954: 186) of *dendritica* to *Placida* Trinchese, 1876 (whose type species [O'Donoghue, 1929: 740] has an albumen gland but unbranched liver diverticula in the cerata [Trinchese, 1879, pl. 15, fig. 1]) is hardly acceptable.

Dr. George M. Moore of Durham, N. H., in a typewritten list (10. III. 1956) which continues his report (1950) on nudibranchs of New England, reported the occurrence of *H. dendritica*. There are no earlier records from the American coast.

Hermaea cruciata Gould (1870: 253) is not identical with *dendritica*. *Hermaea coirala* Marcus (1955a: 108) from the coast of São Paulo may be *cruciata*, but this cannot be decided without *cruciata* from the typical locality —Naushon Island, Mass.

2. *Pleurobranchaea hedgpethi hamva* Marcus, 1955

Occurrence: Point of Cape Hatteras, N. C., 29. VIII. 1958; three specimens. Further distribution of subspecies: Coast of São Paulo, Brazil.

The present specimens were in life lighter colored than the first described ones (Marcus, 1955b: 21). The subspecies differs from *P. hedgpethi hedgpethi* Abbott, 1952, by its dorsally directed flap of the membrane around the genital aperture. In the dissected specimen of *hedgpethi hedgpethi* from the type locality (Port Aransas, Tex.), the spermatheca was much longer than broad. Another *hedgpethi hedgpethi* from Port Isabel Channel, Tex., had a globular spermatheca (Marcus, 1960c), as have the Brazilian and present slugs of *hedgpethi hamva*. Hence the form of the spermatheca varies according to its degree of filling. The slug from Port Isabel Channel (preserved, 45 mm.) had 36 rows with 56 teeth in each half-row of the radula; a Brazilian specimen (preserved, 33 mm.) had 38 and 60 as corresponding numbers; and the two present slugs (preserved, 35 and 32 mm.) have 38 and 33 rows with 70 and 57 teeth. Hence the number of radular teeth lacks systematic significance.

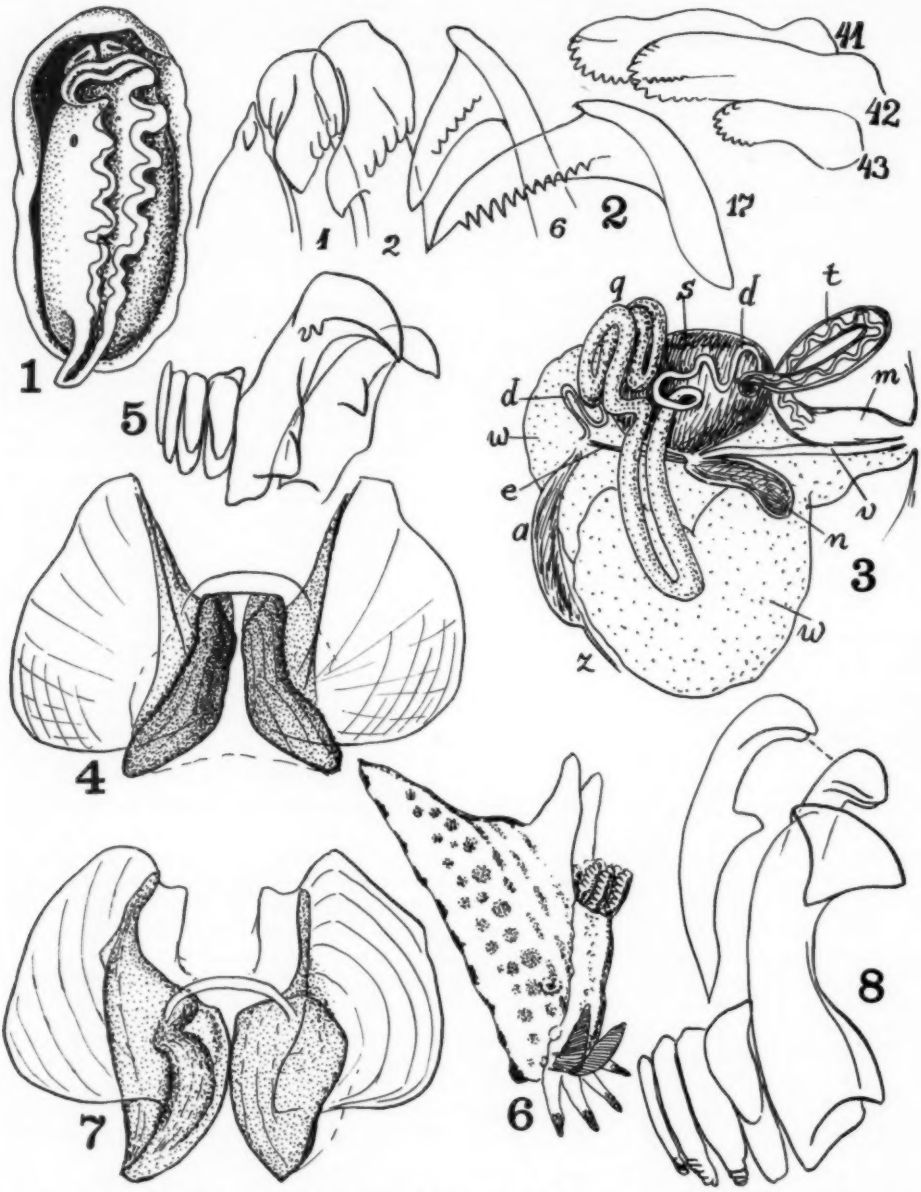
Originally, *Pleurobranchaea hamva* was described as a separate species, but with more detailed knowledge it seems better to consider it as a subspecies.

3. *Chromodoris aila*, spec. nov. (Figs. 1–3)

Occurrence: Beaufort, N. C., 13. VI. 1955; one specimen.

Length 12 mm., breadth 7 mm., height 6 mm. In life, colored as *mörchii* Bergh, 1879, hence red with blue and yellowish marks; in preserved state, uniformly pinkish. Notum smooth, its free edge 2 mm. broad in front and 1 mm. broad behind, without glands. Spicules absent or dissolved. Sole 2 mm. broad, tapering behind, tail standing out beyond notum; anterior border of foot bilabiate with distinct rounded corners.

Tentacles broad, grooved; rhinophores with from 15 to 18 leaves; branchial pouch 3 mm. broad, 2 mm. long, with smooth rim; 13 uni-



FIGS. 1–3. *Chromodoris aila.* FIG. 1. Ventral. FIG. 2 Radula. FIG. 3. Reproductive organs. FIGS. 4–5. *Polycera hummi.* FIG. 4. Jaws. FIG. 5. Radula. FIGS. 6–8. *Polycera chilluna.* FIG. 6. Side view. FIG. 7. Jaws. FIG. 8. Radula.

pinnate gills. Labial grasping ring of bifid hooklets. Radula 57 × 43.1.43. Rhachidian tooth a flat triangle with minute cusp; first lateral tooth with 1 or 2 inner and 4 outer denticles; following teeth with more numerous (up to 16) outer denticles; teeth shortening from thirty-sixth tooth outward, the cups of marginal ones concave, denticulate.

Blood gland over pharynx; salivary glands long, knobby; stomach embedded in digestive gland, only pyloric region free, with ventral caecum. Digestive gland surrounded by ovotestis, except its midventral part.

Hermaphrodite duct (*z*) enters hind end of ampulla (*a*). Sperm duct (*d*) emerges from gland mass (*w*)—first thin, then becomes thick, coiled, prostatic (*q*). Following sections muscular, first thin, then sheathed (*t*)—opening into atrium (*m*). Vagina (*v*) narrow; large spermatheca (s) and narrow spermatocyst (n) neighboring; long insemination duct (*e*) plunges into gland mass near outlet of ampulla.

DISCUSSION. Following Odhner (1957), I use the name *Chromodoris* Alder and Hancock, 1855, for the species with hamate teeth (Marcus, 1960a: 900–901). *Chromodoris aila* has been compared with about forty Caribbean, American Pacific, Atlantic, and Mediterranean species. Except for a few described in the last ten years (White, 1952; Pruvot-Fol, 1953; Marcus 1955a), these species are catalogued in Pruvot-Fol's synopsis (1951b).

4. *Polycera hummi* Abbott, 1952 (Figs. 4–5)

Occurrence: Beaufort, N. C., 12. III. 1956; one specimen. Further distribution: Alligator Harbor, Franklin County, northwest Florida.

As in Abbott's specimens (1952: 3), the velum of the present one bears two pairs of processes; up to four may occur in *P. aurisula* (Marcus, 1960b: 162). The foremost gill is long, but only by degree different from *aurisula*. The same holds for the extra branchial appendages, which are blunt and club shaped in *hummi* and carrot shaped in *aurisula*. The innermost marginal tooth has five small denticles on the cusp in Abbott's material. In the present slug we found the anterior edge of all marginal teeth worn in old rows, and not quite regular in new ones, but always without regular denticles. As in *aurisula* (Marcus, 1960b, fig. 47), the cusps of the two inner marginal teeth project on the inner side. The cusp of the biggest, the second lateral tooth, has one or two denticles, absent in worn rows. Such denticles do not occur in *aurisula*. The jaws of *hummi* (Fig. 4) are 1.9 mm. broad and 1.3 mm. high, not specifically different from those of *aurisula*.

5. *Polycera chilluna*, spec. nov. (Figs. 6–9)

Occurrence: Beaufort, N. C., 11. VII. 1956; one specimen.

Length 18 mm. from velar appendages (3 mm.) to stretched tail (5 mm.), height 7 mm., breadth 6 mm. Colors in life fitting typical pattern of *P. quadrilineata;* light brown when preserved, with interrupted dark marks (Fig. 6). Velar appendages long, pointed, three on left, two on right side (one evidently lost). Velum continues with low ridge on either side to extrabranchial process. Shaft of rhinophores 1 mm., pointed club slanting backward 2 mm. long, containing 25 perfoliations and a small terminal knob. Foot narrow, anterior border simple. Spicules not preserved.

Jaws together 2 mm. broad, 1.4 mm. high, ventroanterior cutting portion brown, dorsolateral arched wings broad, yellow. Radula 13 × 3–4.2.0.2.3–4. Innermost tooth 0.34 mm. with small longitudinal fold on cusp and denticle in middle; second tooth 0.37 mm., cusp with transverse backward fold, base with extended inner angle. Innermost marginal tooth with cusplike anterior edge. Inner upper angles of first to third marginal teeth project over base.

Ampulla (*a*) reniform as in other species of *Polycera* (Marcus, 1956: 52); entrance of hermaphrodite duct (*z*) opposite bifurcation of spermoviduct. Oviduct (*o*) merging into gland mass (*w*). Sperm duct wide, prostatic (*q*), apposed to spermatheca (s), followed by narrow muscular efferent duct (*d*) with glandular swelling. Hook-bearing outer portion projects into deep atrium (*m*). Hooks small, in regular rows. An accessory ganglion (*l*) between male atrium and vagina (*v*). Vagina glandular, entering big spherical spermatheca (*s*) close to origin of long, coiled insemination

Explanation of Letters in Figures

a. ampulla. **b.** penis. **c.** cerebral ganglia. **d.** sperm duct. **e.** insemination (uterine) duct. **f.** fertilization chamber. **g.** abdominal ganglion. **h.** pharynx. **i.** oesophagus. **io.** spermoviduct. **j.** rhinophorial ganglion. **k.** buccal ganglia. **l.** accessory genital ganglion. **m.** male atrium. **n.** spermatocyst. **o.** oviduct. **p.** pedal ganglia. **q.** prostatic part of sperm duct. **r.** anus. **s.** spermatheca (bursa copulatrix). **t.** sheathed part of sperm duct. **u.** valvular apparatus. **v.** vagina. **w.** female gland mass. **x.** salivary glands. **y.** retractors. **z.** hermaphrodite duct.

duct (*e*). This bears longish spermatocyst (*n*) and opens into gland mass near entrance of oviduct (*o*).

DISCUSSION. The color agrees with that of narrow-striped *P. quadrilineata* (Müll.) (Larsen, 1925, col. fig. 6a). That species has much narrower mandibular wings (*ibid.*, text fig. 14) and from 10 to 12 rhinophorial perfoliations (Haefelfinger, 1960: 102; and many others); Berg's indication of up to 18 (1880: 604) is quite exceptional.

Broad wings and about 28 perfoliations (Allan, 1932: 101) occur in *P. capensis* Q. et G., frequent in the growth on ship bottoms (Macnae, 1959: 352–353) and widely distributed. It has one median and two lateral black or red (Macnae) stripes (Barnard, 1927, pl. 19, figs. 7, 8; Allan, 1932, pl. 4, figs. 12, 13; Dakin, 1953, pl. 62), and the spur of the first tooth is near the base. The cusp of the second is narrower than in *chilluna*.

6. *Acanthodoris pilosa* (O. F. Müller, 1776)

Occurrence: Ocean City, Md., 22. V. 1960; two specimens. Further distribution: Mediterranean; Aegean Sea, Palermo; European seas, including western Baltic, Murman Coast, White Sea, Iceland; Greenland; North American Atlantic coast southward to Connecticut; North Pacific southward to Vancouver Island region: northern Japan.

7. *Okenia impexa* Marcus, 1957

Occurrence: Beaufort, N. C., 25. III. 1956; two specimens. Further distribution: Brazil, coast of São Paulo.

8. *Ancula evelinae,* spec. nov. (Figs. 10–14)

Occurrence: Beaufort, N. C., 9. VII. 1955; two specimens.

Length about 3 mm., height 2 mm., breadth 1 mm. In life, cream colored; preserved, pinkish. Nearly symmetrical black blotches: In smaller specimen, one between rhinophores extended to sides of head, reaching gills; a second behind branchiae, bifurcated down to sides; on either side of body, big blotch in middle and small one on root of tail. In larger specimen, all-black areas smaller and each divided in two. Notum smooth, merges into sides without limit. Sole narrow, its borders wavy, probably due to contraction. Tongue-shaped tentacle on each side of mouth. Rhinophores each with 7 or 8 perfoliations and two divergent, finger-shaped appendages in front of their bases. Gills three, bipinnate; on either side one flattened, curved process, longer than branchiae. Tail laterally compressed, with keel.

Labial ring of conical platelets in several irregular rows; outer cones short, blunt; inner ones long, pointed. Radula 40 × 1.1.0.1.1. Inner tooth 21 μ high in oldest rows, outer one absent. Inner tooth of younger rows 52 μ. Its cusp with prominent outer denticle and from 14 to 18 fine ones, base elongated on outer side. Outer tooth claw-shaped, touching base of inner one.

Ovotestis covering digestive gland. Hermaphrodite duct (*z*) rather long, opens into ampulla (*a*) opposite origin of immediately branching spermoviduct. Sperm duct prostatic (*q*), U-shaped. Invaginated efferent duct an acrembolic penis (*b*) with ciliated papilla and hook-bearing collar. Muscular vagina (*v*) leads to longish spermatheca (*s*) and before it enters, gives off long, coiled insemination duct (*e*). Spermatocyst (*n*) with rather long canal. Fertilization chamber (f) lies where insemination duct and oviduct enter into gland mass (*w*). Common genital aperture wide, folded.

The species is named for Mrs. Eveline du Bois-Reymond Marcus. The holotype is the slightly larger, nearly entire slug.

DISCUSSION. Absence of blotches, shorter extrabranchial process, and shape of inner tooth distinguish *A. fuegiensis* Odhner (1926a: 45) from *evelinae*. Color, inner tooth, and several extrabranchial appendages separate *cristata* and *pacifica* (see Bergh, 1881: 631 ff.; MacFarland, 1906: 148). Moreover, *pacifica* has rhachidian teeth behind the first 8 to 10 rows.

9. *Corambella barataríae* Harry, 1953

Occurrence: Beaufort, N. C., and Cape Hatteras, N. C., 30 V. 1956 and 20. VIII. 1959; a total of five specimens. Further distribution: Grand Isle, La. (Harry, 1953); Port Aransas, Tex. (Marcus, 1960c).

10. *Doriopsilla leia,* spec. nov. (Figs. 15–18)

Occurrence: Beaufort, N. C., 20. VII. 1955; three specimens.

Length 7 and 8 mm., breadth 3 mm. Body nearly round, pinkish, soft and smooth. Spicules wanting, possibly dissolved. Free notal edge 0.5–1 mm. broad, with slight radial stripes. Tentacles minute; eight rhinophorial leaves. Genital papilla distinct; anal papilla (*r*) high, to the left of the four tripinnate gills (Pruvot-Fol, 1951a: 40; 1953: 91–92). Foot 1 mm. broad, evenly curved behind and in front, without transverse groove.

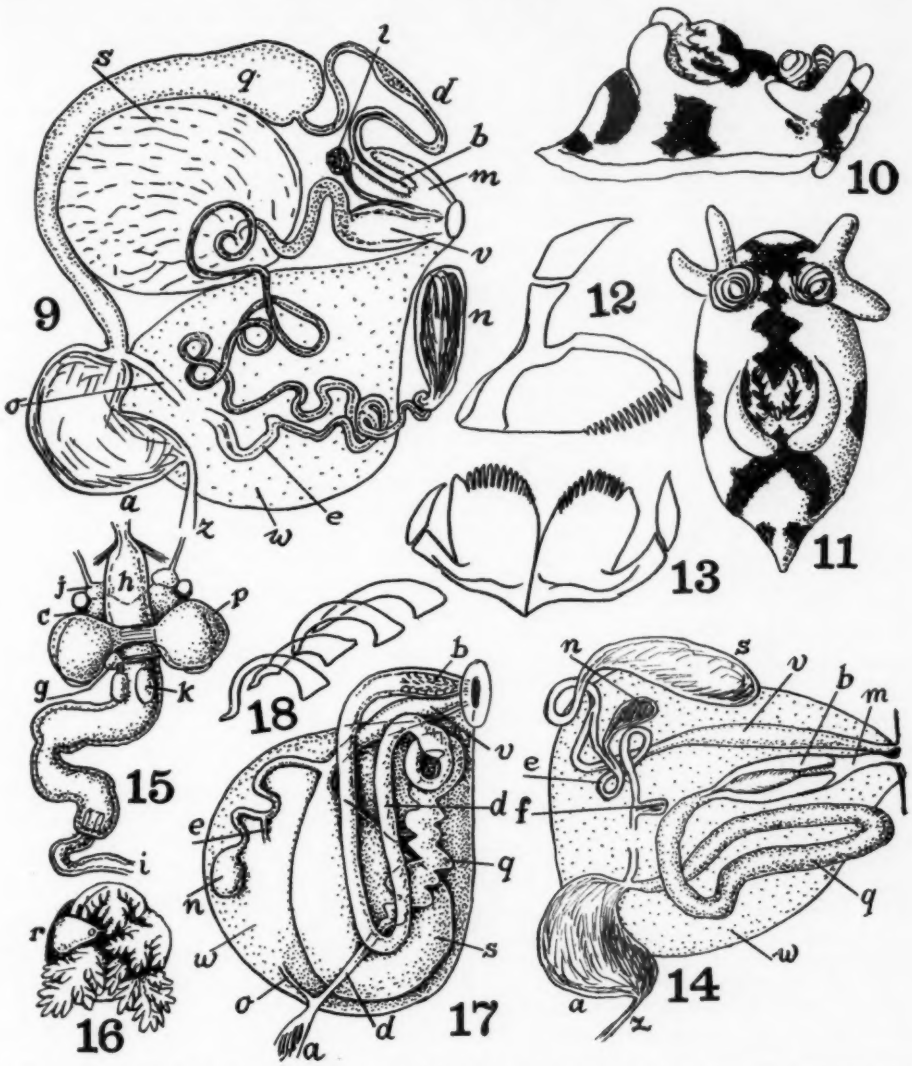


FIG. 9. *Polycera chilluna*, reproductive organs. FIG. 10–14. *Ancula evelinae*. FIG. 10. Lateral. FIG. 11. Dorsal. FIG. 12. Radular row. FIG. 13. Half-row of radula. FIG. 14. Reproductive organs. FIGS. 15–18. *Doriopsilla leia*. FIG. 15. Anterior gut, ventral. FIG. 16. Gills. FIG. 17. Reproductive organs. FIG. 18. Penial hooks.

Eyes black, located close in front of nerve ring, shine through skin behind rhinophores. Nerve ring overlain by coalesced blood glands. Pedal commissure distinct. Buccal ganglia (*k*) close behind nerve ring, ventral to pharynx. Rhinophorial ganglia (*j*) set off from cerebral ones (*c*). Abdominal ganglion (*g*) at right root of viseceral loop.

Inner organs unpigmented; pharynx (*h*) wide; no ptyaline gland. Oesophagus narrow (*i*), can be projected into pharynx (Vayssière, 1901, pl. 3, fig. 19, b). Farther behind, oesophagus widens, forms loop and enters stomach, which is embedded in digestive gland. Small caecum close behind pylorus. Gut fastened by muscles.

Spermoviduct bifurcates near outlet of ampulla (*a*). Oviduct (*o*), to the left, enters voluminous gland mass (*w*). Sperm duct (*d*), to the right and forward—first narrow, straight, then wide, coiled, prostatic (*q*). Following loop (*d*), muscular, enters short penial pouch formed by atrium. Here, efferent duct ends as invaginated penis (*b*) with many colorless hooks. Muscular vagina (*v*) leads to large, pyriform spermatheca (*s*). At its entrance sinuous duct to spermatocyst (*n*) begins. Insemination canal (*e*) goes out from this duct.

The holotype is one of the 8-mm.-long slugs.

DISCUSSION. The position of the buccal ganglia is the disjunctive character of the genus. Muscles which move the anterior gut must not be confounded with cerebro-buccal connectives (Eliot, 1906a: 366). Pruvot-Fol (1951a: 39; 1952: 413; 1954: 336) stressed absence of ptyaline gland in *Doriopsilla*, but Bergh (1896: 457) described such for the type species, *D. pallida* (1902: 194) and *D. capensis* (1907: 64). *Doriopsilla laevis* Bergh, 1905, can be separated from *leia* by its dorsal skin glands, 0.12 mm. in diameter. The other previously described species of *Doriopsilla* have dorsal granules or tubercles.

11. *Doriopsilla pharpa*, spec. nov. (Figs. 19–21)

Occurrence: Beaufort, N. C., 20. VII. 1955, VII. 1956, 25. IV. 1960; a total of six specimens.

Length from 4 to 17 mm. In life, orange with brown specks produced by chromatophores in connective tissue, or bright yellow. Biggest mature slug 9 mm. broad, 4.5 mm. high; free edge of notum 2 mm. broad; genital aperture 5 mm. behind fore end. Body flat, firm; notum with irregular papillae stiffened by bundles of small spicules, 15 μ in diameter. In connective tissue spicules larger: 1.2 mm. long, 70 μ in diameter, disposed diagonally, crossing at right angles. Rim of branchial and rhinophorial pockets smooth. Tentacles small; rhinophores with twelve perofilations. Foot rounded in front, pointed behind. Eyes, four gills, and anal papilla as in *D. leia*.

Pedal ganglia (*p*) contiguous; buccal ganglia (*k*) touch nerve ring; salivary glands (*x*) attached directly to hind end of foremost wide (pharyngeal) part of gut. No ptyaline gland. Oesophagus (*i*) a long loop; digestive gland covers stomach; pylorus with small caecum.

Genital organs essentially as in *D. leia*. Long ampulla (*a*) with hermaphrodite duct (*z*) and spermoviduct (*io*) at opposite ends. Spermoviduct divides a certain distance from ampulla. Sperm duct (*d*) begins narrow, continues as widened prostatic loop (*q*) and ends as eversible hooked penis (*b*). Hooks touch one another. Vagina (*v*) begins very wide, then narrows and enters pyriform spermatheca (*s*). At its entrance, duct to spermatocyst (*n*) emerges. Insemination duct (*e*) enters gland mass (*w*), approaching oviduct (*o*).

DISCUSSION. This species differs from *D. leia* by the possession of dorsal tubercles and contiguous pedal ganglia. Distinguishing characters of other valid species of *Doriopsilla*:

albopunctata (Cooper, 1863); synonym: *reticulata* (Cockerell and Eliot, 1905). Rhinophores with 30 leaves.

miniata (Alder and Hancock, 1864). Back with whitish reticulum.

areolata Bergh, 1880. Type species. Whitish network on notum; 25 rhinophorial perfoliations.

pelseneeri Oliveira, 1895. Dorsal tubercles enormous, 8 per cent of body length.

pallida Bergh, 1902. Ptyaline gland.

laevis Bergh, 1905. Cutaneous glands, 0.12 mm. in diameter.

capensis Bergh, 1907. Ptyaline gland; six gills.

unnamed species White, 1951. Eleven branchiae.

rarispinosa Pruvot-Fol, 1951. Penial hooks widely interspaced.

pusilla Pruvot-Fol, 1951. Four thick tubercles in two rows.

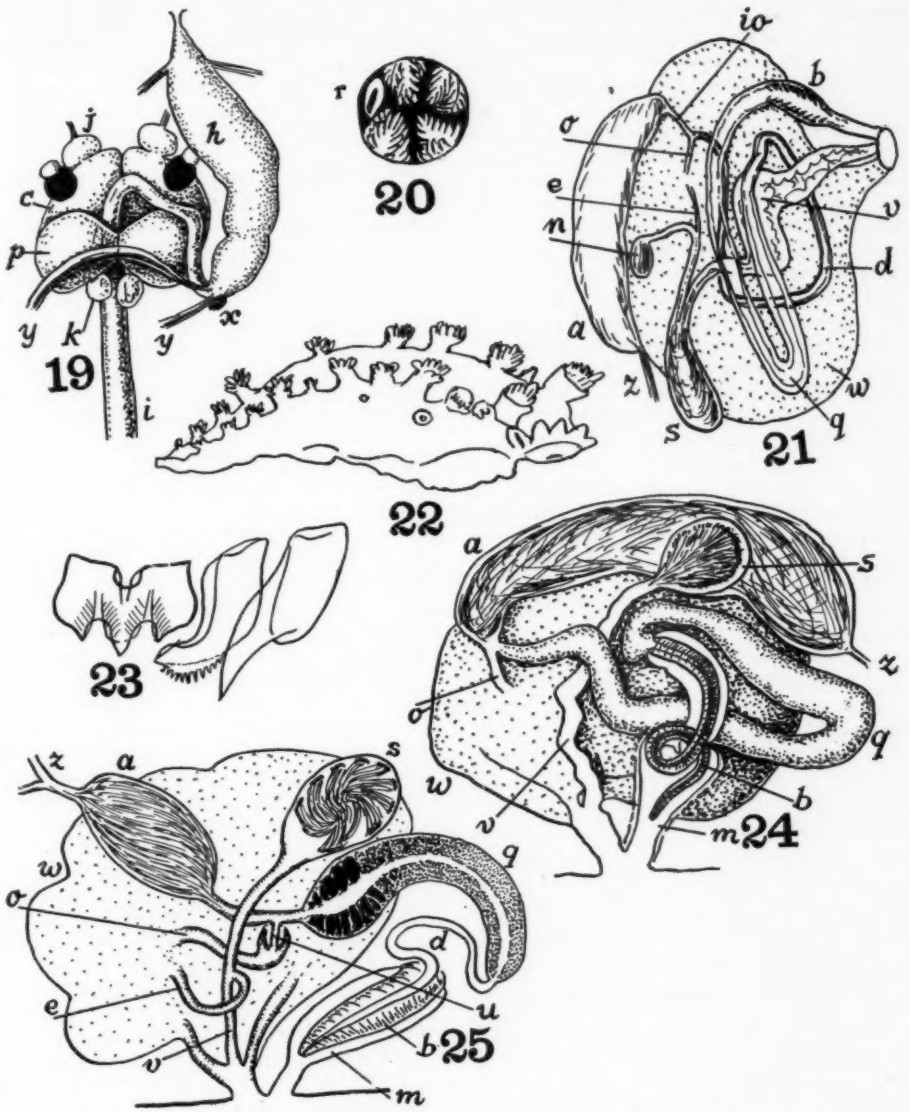
fedalae Pruvot-Fol, 1953. White net between tubercles; penial hooks absent or minute.

12. *Tritonia* (*Tritonidoxa*) *wellsi*, spec. nov. (Figs. 22–24)

Occurrence: Beaufort, N. C., 9. VII. 1956; three specimens.

Length 5, 6, and 7 mm., height of last specimen 3 mm., breadth of notum and sole 2 mm. In life, inner organs orange or salmon, showing through body wall; gills translucent whitish. Back and sides smooth; hind end tapers. Genital papilla broad, under second or third branchial tuft; anus near fourth. Veil small, not bilobed, with six appendages, the two outermost of which are spoon-shaped tentacles. Rhinophorial sheaths high with smooth borders. Edge of notum webbed between bases of tenth and eleventh gills. Fully developed branchiae of rather uniform size and simply ramified.

Masticatory border of jaw with about eight rows of pointed denticles, quincuncial uniform cones, larger in middle than on margins. Radula 21 × 11.1.1.1.11. Median tooth tricuspidate,



FIGS. 19–21. *Doriopsilla pharpa*. FIG. 19. Anterior gut, ventral. FIG. 20. Gills. FIG. 21. Reproductive organs. FIGS. 22–24. *Tritonia* (*Tritonidoxa*) *wellsi*. FIG 22. Side view. FIG. 23. Middle of radula. FIG. 24. Reproductive organs. FIG. 25. *Miesea evelinae*, reproductive organs.

lateral teeth hook-shaped. Innermost lateral tooth hollowed on inner side, cusp with about twelve denticles.

Ampulla (*a*) distended, farthest to the left. Outlet bifurcates; oviduct immediately enters gland mass (*w*); sperm duct prostatic (*q*) on entire course. Glandular sperm duct attains fundus of long sac, a penial pouch, set off from atrium (*m*) by fold. Penis (*b*) muscular, flagelliform, curled, 1 mm. long, 30–40 μ in diameter; projects into atrium. Ectal parts of vagina (*v*) and nidamental duct in common.

Vagina about 0.7 mm. long leads into globular spermatheca (*s*) (diameter: 0.35 mm.). Here spermatozoa fastened to wall with heads.

This species is named for Dr. Harry W. Wells. The holotype is the 6-mm. long, entire slug.

DISCUSSION. It is advisable to use the same term for the ectal (in most cases sole) "seminal vesicle" (Bergh) of Dendronotacea and the always single one of Eolidacea. The spermatheca has a short duct (vagina) in the type species of *Duvaucelia* Risso, 1828 (Odhner, 1939: 43), and a long one in that of *Tritonia* Cuvier, 1797 (Odhner, 1926b, fig. 12). The later name (Pruvot-Fol, 1931: 314, 747–748) is generally accepted. Evidently in correlation with the flagelliform penis (Odhner, 1926b: 17) the vagina is still longer in *Tritonidoxa* Bergh, 1907 (Odhner, 1926a: 32–33, emend.). In *T. capensis* Bergh (1907: 87) (the type species of *Tritonidoxa*) and *T. ingolfiana* (Bergh, 1900: 19) the masticatory border is smooth; in *griegi* Odhner (1922: 6, 8) and *wellsi*, this border is denticulate. Smooth cusp of intermediate tooth separates *griegi* from *wellsi*.

13. *Scyllaea pelagica* Linné, 1758

Occurrence: Beaufort, N. C., VII. 1956; one specimen.

Further distribution: Pelagic in warm and warm-temperate seas, clinging to floating seaweeds, chiefly *Sargassum*, feeding on hydroids, and also swimming in open sea. In the Atlantic occasionally northward to the British Isles (Eliot, 1910: 163) and Woods Hole region, southward to South Africa. In the Indo-West Pacific from Red Sea to middle Japan and New Caledonia. One record from eastern Pacific without locality (Bergh, 1894: 145).

14. *Miesea evelinae* (Marcus, 1957) (Fig. 25)

Occurrence: Beaufort, N. C., 1. VIII. 1955; one specimen. Further distribution: Brazil, coast of São Paulo.

The species described as *Embletonia evelinae* Marcus (1957: 466) has since then been found also 350 km. southwest from the original locality. It must be transferred to the Dotonidae and becomes the type species of the new genus *Miesea*.

***Miesea*, gen. nov.**

Dotonidae without sheaths of the rhinophores; cerata simple, without gills; veil small; radula 0.1.0.

In sections of some Brazilian specimens the shafts of the rhinophores show a fold around their basal third, but no sheath into which the club can be retracted. In some preserved animals the skin of the cerata is somewhat shrunken around the racemose digestive gland, so that an illusion of bosses is produced.

Independently of systems of classification used for the Nudibranchia, such as that of Odhner considering rhinophorial sheaths "an essential characteristic of the Dendronotacea" (1934: 231; 1936: 1060), or that of Pruvot-Fol (1954: 402, 410) separating Dotoinae and Embletoninae as subfamilies of the Dotoidae, this species would at first sight be allocated to *Embletonia*.

The veil and the at most vestigial tentacles separate the Embletoniidae from the Eolidacea (Odhner, 1939: 50), but their radulae are euthonidan. The main duct of the digestive gland is dorsal to the ovotestis, as in most eolidacean nudibranchs (Bergh, 1892: 1012–13). In *Doto* the gonad lies over the main liver duct (Alder and Hancock, 1846: fam. 3, pl. 4; Odhner, 1922: 37; Pruvot-Fol, l.c.). *Miesea evelinae* has this dotoidan topography. The generative organs (Fig. 25) with unarmed penis (*b*) and ciliated valve (*u*) at bifurcation of spermoviduct agree in *Doto* and *Miesea*. In *Embletonia* and *Tenellia* the penis is armed, and the valve described of *Tenellia fuscata* is heavily muscular (Chambers, 1934: 621).

Anterior glands around mouth; a big, left-sided posterior gland; salivary glands; extremely delicate jaws; and radula with slanting, converging rows brought about by variable position of lateral denticles are characters which *Miesea* has in common with *Doto*.

15. *Catriona tina* Marcus, 1957

Occurrence: Beaufort, N. C., 1. VIII. 1955; one specimen. Further distribution: Brazil, coast of São Paulo; Virginia Key, Miami, Fla.

16. *Dondice occidentalis* (Engel, 1925)

Occurrence: Beaufort, N. C., 12. VI. 1956; two specimens. Further distribution: Brazil, coast of São Paulo; Jamaica; Virginia Key, Miami, Fla.

17. *Cratena kaoruae* Marcus, 1957

Occurrence: Beaufort, N. C., 30. V. 1955; Cape Hatteras, N. C., 12. II. 1959; two specimens. Further distribution: Brazil, coast of São Paulo; coasts of Texas and Louisiana.

18. *Aeolidia papillosa* (Linné, 1767)

Occurrence: Ocean City, Md., 22. V. 1960; two specimens. Further distribution: White

Sea (Mileikovsky, 1960: 668); North Atlantic, southward to Portugal, possibly to the Mediterranean, on the American coast southward to Rhode Island; North Pacific, from Alaska to Southern California, on the Asiatic coast to Sakhalin and east coast of Hokkaido. Specimens from South America, Valparaiso to Falkland Islands, may perhaps be separable as var. *serotina* Bergh, 1874.

19. *Baeolidia benteva* Marcus, 1958

Occurrence: Cape Hatteras, N. C., 8 X. 1959; three specimens. Further distribution: Brazil, coast of São Paulo.

20. *Berghia coerulescens* (Laurillard, 1830)

Occurrence: Beaufort, N. C., 30. V. 1956; one specimen. Further distribution: Mediterranean; East Atlantic, northward to French Channel, southward to Morocco; West Atlantic: Brazil, coast of São Paulo; West Indies; coast of Texas.

Zoogeographic Remarks

To the twenty above-mentioned species can be added *Aplysia morio* Verrill, 1901, and *Aplysia willcoxi* Heilprin, 1886, from Beaufort, N. C., identified by Dr. Harry W. Wells. The former ranges from Rhode Island to Texas and Bermuda (Eales, 1960: 328), and the latter from Woods Hole to Guadeloupe (*ibid.*, 350). Six species from North Carolina are new. These and the pelagic species, *Scyllaea pelagica*, cannot be analyzed zoogeographically. Of the remaining species and subspecies, two eurythermal boreo-arctic ones—*Acanthodoris pilosa* and *Aeolidia papillosa*—have their range extended on the North American east coast from the Cape Cod region to Maryland. This is not surprising with regard to their Mediterranean (*A. pilosa*) and western American (*A. papillosa*: Laguna Beach, Calif.) occurrences.

Contrary to these species whose ranges in both northern temperate oceans are connected by the Arctic, the distribution of the non-arctic species, *Hermaea dendritica*, cannot be understood in the present state of knowledge. Its transatlantic distribution leads to the supposition that the Atlantic Ocean in Tertiary times was narrower than today. Its occurrence in southern and middle Japan makes probable the possibility that it also lives in the Red Sea and the Indian Ocean; hence it exhibits a Tethyan distribution. The species feeds chiefly on *Codium*, which does not belong to the ordinary growth on ship bottoms. Hermaeids are easily overlooked by collectors (Macnae, 1954: 60), so that the gap in the range of the temperate-water species *Hermaea dendritica* is certainly due to the present state of knowledge. This gap cannot be filled by uniting *H. capensis* Macnae (1954: 59) and *H. aoteana* Powell (1937: 123) with *dendritica*, though these species from Cape Province and Auckland, respectively, have *dendritica*-like appearances. Their radulae, however, are different, while that of the Japanese *dendritica* (Baba, 1955: fig. 9) agrees with the European one (Pruvot-Fol, 1954: fig. 71, i).

The range of *Berghia coerulescens* could, theoretically, have been brought about by drifting logs bearing hydroids. The larva has a shell (Trinchese, 1881, pl. 78, figs. 6–8) of Thorson's type B (1946: 268), small velum, and no eyes, hence is very poorly equipped for planktonic life (*ibid.*, 270). It is possible that such larvae hatched from eggs laid on the raft settle on it a few hours later; the slugs themselves hardly live for more than one year. Transatlantic distribution of such short-lived nudibranchs is better explained by a former narrower Atlantic Ocean than by the raft hypothesis.

Of the analyzable species from North Carolina (*Aplysia morio* and *A. willcoxi* included), 69 per cent are West Atlantic warm-water elements. Only *Polycera hummi* and *Corambella barataríae* seem to be restricted to the southeast coast of the United States. Both have a range disjunct by a break in southern Florida, as do Hedgpeth's examples *Littorina irrorata* and *Sesarma cinereum* (1953, figs. 43, 44). It is not known whether this break also applies to the rather eurythermal *Aplysia morio*.

Of the analyzable species from North Carolina, 62 per cent are also known from southern middle Brazil. This is less than the corresponding 72 per cent found in a collection from Miami (Marcus, 1960b). The available species are relatively few in number, and the compared collections cover small areas. However, they indicate a certain uniformity of the intertidal opisthobranch fauna between northern and southern tropic waters which decreases northward.

LITERATURE CITED

ABBOTT, R. T. 1952. Two New Opisthobranch Mollusks Belonging to the Genera *Pleurobranchaera* and *Polycera*. Fla. State Univ. Stud. 7.

ALDER, J., AND A. HANCOCK. 1846. A Monograph of the British Nudibranchiate Mollusca 2. Ray Society: London. 13 pls.

ALLAN, J. K. 1932. Australian nudibranchs. Austral. Zool. **7** (2): 87-105.

BABA, K. 1955. Opisthobranchia from Sagami Bay. Supplement. Iwanami Shoten, Tokyo. 59 pp., 20 pls.

BARNARD, K. H. 1927. South African nudibranch Mollusca, etc. Ann. S. Afr. Mus. **25**: 171–215.

BERGH, R. 1874. Beiträge zur Kenntniss der Aeolidiaden. I. Verh. zool. bot. Ges. Wien **23**: 596–628.

———. 1880. Beiträge zu einer Monographie der Polyceraden. I. Verh. zool. bot. Ges. Wien **29**: 599–652.

———. 1881. Beiträge zu einer Monographie der Polyceraden. II. Verh. zool. bot. Ges. Wien **30**: 629–668.

———. 1892. Malacologische Untersuchungen. In C. Semper (ed.), Reisen im Archipel der Philippinen **3** (18): 995–1165.

———. 1894. Die Opisthobranchien. Bull. Mus. Comp. Zool. Harvard **25** (10): 125–233.

———. 1896. Ueber die Gattung *Doriopsilla*. Zool. Jahrb. Syst. **9** (3): 454–458.

———. 1900. Nudibranchiate Gastropoda. Danish Ingolf Exped. **2** (3): 1–49. Copenhagen.

———. 1902. Gasteropoda opisthobranchiata. Danske Vid. Selsk. Skr. ser. 6. Naturvid. Afd. **12** (2): 159–218 (1–60).

———. 1907. The Opisthobranchiata of South Africa. Trans. S. Afr. Phil. Soc. **17** (1): 1–144.

CHAMBERS, L. A. 1934. Studies on the organs of reproduction in the nudibranchiate molluscs. Bull. Amer. Mus. Nat. Hist. **66**: 599–642.

COCKERELL, TH. D. A., AND CH. ELIOT. 1905. Notes on a collection of Californian nudibranchs. Jour. Malacol. **12**: 31–53.

DAKIN, W. J. 1953. Australian Seashores. Angus & Robertson: Sydney & London.

EALES, N. B. 1960. Revision of the world species of *Aplysia*. Bull. Brit. Mus. (Nat. Hist.) **5** (10): 268–404.

ELIOT, CH. 1960a. The genus *Doriopsilla*. Jour. Conchol. **11**: 366–367.

———. 1960b. Report upon a collection of Nudibranchiata from the Cape Verde Islands. Proc. Malacol. Soc. Lond. **7**: 131–159.

———. 1910. A Monograph of the British Nudibranchiate Mollusca 8. Ray Society: London. 198 pp., 8 pls.

FISCHER, P. 1887. Manuel de Conchyliologie. F. Savy: Paris.

GOULD, A. A. 1870. Report on the Invertebrata of Massachusetts. 2d ed. (Mollusca, W. G. Binney). Wright & Potter: Boston, Mass.

HAEFELFINGER, H. R. 1960. Beobachtungen an *Polycera quadrilineata*. Rev. Suisse Zool. **67** (3): 101–117.

HARRY, H. W. 1953. *Corambella baratariae*, a New Species of Nudibranch from the Coast of Louisiana. Occ. Pap. Mar. Lab. Louisiana State Univ. 8.

HEDGPETH, J. W. 1953. An introduction to the zoogeography of the Northwestern Gulf of Mexico with reference to the invertebrate fauna. Publ. Inst. Mar. Sci. Univ. Texas **3** (1): 107–224.

HOFFMANN, H. 1932–39. Opisthobranchia. I. Bronn, Kl. Ordn. 3, II: Buch 3. Akadem. Verlagsges. Leipzig.

JOHNSON, CH. W. 1934. List of marine Mollusca of the Atlantic coast from Labrador to Texas. Proc. Boston Soc. Nat. Hist. **40**: 1–204.

LARSEN, M. 1925. Nudibranchfaunaen i Dröbaksundet. II. Skr. Norske Vid. Akad. Oslo. Naturv. Kl. **2**: 1–60.

MACFARLAND, F. M. 1906. Opisthobranchiate Mollusca from Monterey Bay, California, and vicinity. Bull. U. S. Bur. Fish. **25**: 109–151.

MACNAE, W. 1954. On four saccoglossan molluscs new to South Africa. Ann. Natal Mus. **13** (1): 51–64.

———. 1959. The families Polyceridae and Goniodorididae in Southern Africa. Trans. R. Soc. S. Afr. **35** (4): 341–372.

MARCUS, E. 1955a. Opisthobranchia from Brazil. Bol. Fac. Fil. Univ. S. Paulo, Zool. **20**: 89–263.

———. 1955b. Sea-hares and side-gilled slugs from Brazil. Bol. Oceanogr. S. Paulo 6 (1-2): 3–49 (published 1957).

———. 1956. Notes on Opisthobranchia. Bol. Oceanogr. S. Paulo **7** (1–2): 31–80 (published 1958).

———. 1957. On Opisthobranchia from Brazil (2). J. Linn. Soc. London, Zool. **43** (292): 390–486.

———. 1958. On Western Atlantic Opisthobranchiate Gastropods. Amer. Mus. Novit. No. 1906: 82 pp.

———. 1960a. Opisthobranchia aus dem Roten Meer und von den Malediven. Akad. Wiss. Lit. Mainz, Math. Nat. Kl. 1959, no. 12: 871–934.

———. 1960b. Opisthobranchs from American Atlantic warm waters. Bull. Mar. Sci. Gulf & Carib. **10** (2): 129–203.

———. 1960c. Opisthobranchs of the northwestern Gulf of Mexico. Publ. Inst. Mar. Sci. Univ. Texas **6** (1959): 251–264.

MILEIKOVSKY, S. A. 1960. On the relation between spawning range of a species and its zoogeographic belonging in marine invertebrates. Akad. Nauk SSSR, Zool. Gurn. **39** (5): 666–669.

MOORE, G. M. Progress report on investigations of the Nudibranchiata of New England. Biol. Bull. **99**: 352–353.

ODHNER, N. HJ. 1907. Northern and arctic invertebrates in the collection of the Swedish state museum. III. K. Svenska Akad. Handl. **41** (4): 1–118.

———. 1922. Norwegian opisthobranchiate mollusca in the collection of the zoological museum of Kristiania. Nyt Mag. Naturvid. **60**: 1–47.

———. 1926a. Die Opisthobranchien. Further zool. res. Swed. Antarct. Exped. **2** (1): 1–100.

———. 1926b. Nudibranchs and lamellariids from the Trondhjem Fjord. Kgl. Norske Vidensk. Selsk Skr. 1926. **2**: 1–36.

———. 1934. The Nudibranchiata. Brit. Antarct.

("Terra Nova") Exp. Brit. Mus. Zool. **7** (5): 229–309.

———. 1936. Nudibranchia Dendronotacea, a revision of the system. Mém. Mus. Hist. Nat. Belgique sér. 2, **3**: 1057–1128.

———. 1939. Opisthobranchiate Mollusca from the western and northern coasts of Norway. Kgl. Norske Vidensk. Selsk. Skr. 1939, **1**: 1–93.

———. 1957. *Chromodoris* contra *Glossodoris*. A systematic-nomenclatorial controversy. Proc. Malacol. Soc. Lond. **32**: 250–253.

O'DONOGHUE, CH. H. 1922. On the family Doriopsidae (Doridopsidae). Proc. Malacol. Soc. Lond. **15** (2–3): 142-145.

———. 1929. Report on the Opisthobranchia. Trans. Zool. Soc. Lond. **22** (6): 713–841.

POWELL, A. W. 1937. New species of nudibranchiate Mollusca from Auckland waters. Rec. Auckl. Inst. Mus. **2** (2): 119–124.

PRUVOT-FOL, A. 1931. Note de systématique sur les opisthobranches. Bull. Mus. Hist. Nat. sér. 2, **3**: 308–316, 746–755.

———. 1951a. Études des nudibranches de la Méditerranée (2). Arch. Zool. Expér. Génér. **88**: 1–80.

———. 1951b. Révision du genre *Glossodoris* Ehrenberg. Jour. Conchyl. **91**: 76–164.

———. 1952. Compléments à la connaissance de *Doriopsilla areolata* Bergh. Bull. Soc. Zool. France **77** (5–6): 410–414.

———. 1953. Étude de quelques opisthobranches de la côte Atlantique du Maroc et du Sénégal. Trav. Inst. Sci. Chérif. no. 5, sér. zool. **2**: 1–105.

———. 1954. Mollusques opisthobranches. Faune de France **58**: 1–460. Paul Lechevalier, Paris.

THORSON, G. 1946. Reproduction and larval development of Danish marine bottom invertebrates, etc. Meddel. Kommo. Danmarks Fish. & Havunders. ser. Plankton **4** (1): 1–523.

TRINCHESE, S. 1877–79. Aeolidiae e famiglie affini del porto di Genova. Atti Univ. Genova **2**: 1–94.

———. 1881. Per la fauna marittima Italiana: Aeolididae, etc. Atti Accad. Lincei, Mem. Cl. Sci. Fis. ser. 3, **11**: 3–142.

VAYSSIÈRE, A. 1901. Recherches zoologiques et anatomiques sur les mollusques opisthobranches, etc. III. Ann. Mus. Hist. Nat. Marseille, Zool. **6**: (1): 1–130.

WHITE, K. 1952. On a collection of molluscs from Dry Tortugas. Proc. Malacol. Soc. Lond. **29** (2–3): 106–120.

AN EXPERIMENTAL ANALYSIS OF FACTORS AFFECTING THE DEVELOPMENT OF ADRENAL GLANDS WITH SPECIAL REFERENCE TO *AMBLYSTOMA PUNCTATUM*[1]

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It is generally accepted that the adrenal glands of adult vertebrates are composed of two parts, each arising from a different embryonic germ layer. The adrenal medulla is known to arise from ectoderm, especially the neural crest, while the cortex has been shown to originate from the intermediate mesoderm. The latter component appears first and is invaded by the former. The extent to which this invasion takes place varies in each of the vertebrate classes. In elasmobranchs, the cortical tissue, referred to as interrenal bodies, is separated from the medullary part, called the suprarenal or chromaffin bodies. In mammals the invasion of the future medullary cells into the adrenal cortical primordia is so great that they eventually become surrounded by cortex, which continues to proliferate around them.

Normally mammalian adrenal tissue is confined to two discrete glands, one lying anterior to each kidney. In the intervening classes between elasmobranchs and mammals, there are examples of varying degrees of proximity and juxtaposition of the two adrenal components. Reference is frequently made to the anomalous condition reported in man and other mammals of accessory adrenal nodules along the inferior vena cava and aorta. Nodules have been found which contain either both adrenal components or only one component. (Nelson, 1939; Lasher, 1947; Graham, 1953.) This type of gland, although abnormal in mammals, normally occurs in urodeles. Figure 1 shows the location of adrenal tissue along the walls of the vena cava and efferent renal veins in an adult *Amblystoma punctatum*. In amphibia, as well as in mammals, the normal inherited pattern of behavior of the adrenal components can be

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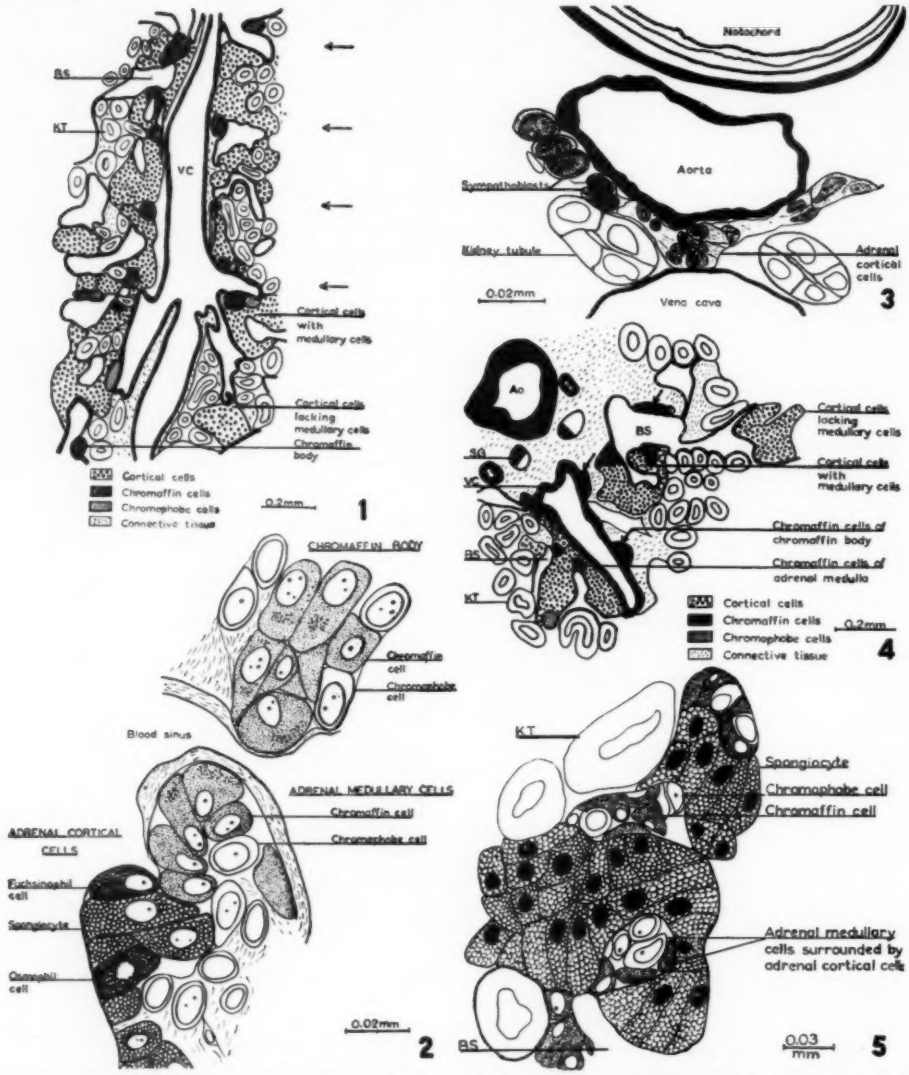


FIGS. 1–5. Adrenals of *Amblystoma punctatum*. FIG. 1. Frontal section of kidneys of 170-mm. adult. Arrows indicate the pseudo-segmental appearance of chromaffin tissue. FIG. 2. Adrenal cells and a chromaffin body of 170-mm. adult. FIG. 3. Young adrenal cortical lobule in 26-mm. larva. FIG. 4. Transverse section of kidney of 170-mm. adult. Arrows indicate location of chromaffin bodies. FIG. 5. Frontal section of adrenal tissue of 170-mm. adult. AO, aorta; BS, blood sinus of efferent renal vein; KT, kidney tubule; SG, sympathetic ganglion; VC, vena cava.

altered. By growing larvae in water containing estradiol, Witschi (1951b), Segal (1953), and Chang and Witschi (1955) were able to stimulate the development of mammalian-like adrenals in frogs. The largest adrenal masses were anterior to the kidneys, where hyperplasia of cortical tissue was so great that it completely surrounded the adrenal medullary cells. Previous investigations have thus demonstrated alterations of the normal inherited pattern of adrenal morphogenesis. The primary purpose of the present investigation was to determine the

factors which influence the morphogenesis of the adrenal glands in *Amblystoma punctatum*.

Three aspects of adrenal development were studied in *Amblystoma punctatum*. First, normal development from the earliest appearance of the adrenal primordia through metamorphosis to the definitive adult organs was investigated as a basis for further experimental study. Second, operations involving the removal of trunk neural crest were performed. Third, trunk neural crest was grafted on the ventral sides of *Amblystoma punctatum* larvae which had recovered from a previous operation for the removal of their own trunk neural crest. It was expected that the experiments would provide information regarding the factors which bring about the union, differentiation, and proliferation of the embryonic cells from widely separated regions which form the adrenal complex. It was also hoped that this series of experiments would provide information on the influence of the migration pathway on the differentiation of future adrenal medullary cells.

Materials and Methods

Eggs of *Amblystoma punctatum* were obtained at pre-neurula stages from temporary ponds in the vicinity of Chapel Hill and Greensboro, North Carolina. In the laboratory, both controls and experimental animals were kept in a constant temperature room at 12°C. For the study of normal development, *Amblystoma punctatum* larvae were taken from the ponds for immediate fixation at each stage of development used in this study. There was no observable difference between field and laboratory control animals. After stage 48, larvae were fed brine shrimp nauplii. From mid-larval life to metamorphosis, they received worms (*Enchytraeus albidus*). *Amblystoma punctatum* adults were obtained from wooded areas near Hyde Park, New York, and from Quivira Specialties Company, Topeka, Kansas.

Histological series of normal developmental stages of the adrenal glands were made of *Amblystoma punctatum* from larval stage 46 (10 mm.) to 175-mm. mature adults. In larval stages the entire animal was sectioned. In adults the kidneys were excised before fixation. All tissue was cut at 6 μ. Camera lucida drawings with accompanying ocular micrometer measurements were made of all significant stages in normal adrenal development. The following combinations of fixatives and staining techniques were used:

1. Fixative: Champy's fluid
   Stain: Dawson and Friedgood methods for pituitary cells (modified)
   Special Staining: Cytoplasm selectively stained in:
   a. Chromaffin cells
   b. Osmophil cells
   c. Acidophil cells of adrenal cortex
2. Fixative: Ludford's osmic acid fixative
   Stain: Altmann's acid fuchsin
   Special Staining: Cytoplasm selectively stained in:
   a. Osmophil cells
   b. Fuchsinophil cells
3. Fixative: Bouin's solution
   Stain: Ehrlich's haematoxylin and light green
   Special Staining: Nucleus and cytoplasm clear in:
   a. Medullary cells (no chromaffin reaction)
   b. Spongiocytes

The first method used was the only one in which both osmophilic and chromaffin cells were adequately demonstrated in the same sections. A modification of the method used for *Rana pipiens* by Stenger and Charipper (1946) was made for urodeles. The modification involved extending both the fixation and postchroming time to two days. After only one day of fixation in Champy's fixative, the osmophil granules showed signs of dissolving in the xylols, alcohols, or mounting media (balsam, oil of cedar, oil of cloves, which contain fat solvents). After only one day of postchroming the chromaffin cells were a light yellow-brown which was not clearly defined.

Normal Development of Adrenals

The normal development of the adrenals of *Amblystoma punctatum* has not been thoroughly investigated at all developmental stages. Branin (1937), who investigated adrenal cortical develpoment in *Hemidactylium scutatum*, included a summary of the few similar investigations in urodeles. He did not investigate the origin and development of the adrenal medullary cells. One obstacle to the investigation of both medullary and cortical components is the fact that different histological methods of fixation and staining are required for clear demonstration of each cell type (Fig. 2). In the adrenal cortex the osmophil cells contain osmophil granules known to be the site of adrenal cortical hormones. The term spongiocyte designates the vacuolate osmophil cell, which has lost osmophil granules. Fuchsinophil cells are smaller than

osmophil cells and contain fuchsinophil granules, which are acidophilic.

Literature on the development of the adrenal glands contains many direct and indirect implications that an inductive sequence might exist and account for the relationship between the two adrenal components with different germ-layer derivation. To investigate this possibility, special attention was given in the normal development of *Amblystoma punctatum* adrenal tissue to an analysis of the following points: origin, migration pathways, adjacent tissue, and differentiation. Previous investigations have concentrated on the development of one component. The present investigation focused on the interrelations of the two adrenal components throughout larval and adult life.

ORIGIN OF ADRENAL CORTICAL CELLS. It has been demonstrated that the first adrenal component to appear in the mesonephric region of the salamander is the adrenal cortical anlagen (Branin, 1937). The place of origin is controversial. In the present study they were first observed after feeding (10–12 mm.) between the archinephric duets and the aorta. They consisted of clusters of from three to five small cells in the region between the pronephroi and the mesonephroi (Fig. 3). At this formative stage it was difficult to distinguish between mesonephric anlagen and adrenal cortical anlagen. They appeared to originate from the same mass of cells. At a slightly later stage, the adrenal cortical primordia were found to differ from the mesonephric anlagen in the following two respects:

1. The adrenal cortical primordia adhered closely to the dorsal and dorsolateral walls of the vena cava while the mesonephric primordia did not make contact with the vena cava at this stage.

2. The developing adrenal cortical cells adhered to each other in compact masses. In contrast, the nephric cells soon organized into tubules, forming a lumen.

The appearance of additional adrenal cortical anlagen was observed to take place in an anterior-posterior time sequence throughout larval life. At later larval stages, from 20 mm., the origin of the adrenal cortical primordia was clearly distinguishable. In the same animal the more mature anterior cortical lobules at the level of the liver showed histological differentiation; at corresponding locations in the mesonephroi at progressively more posterior levels, the cortical primordia showed less and less differentiation and approached more and more closely the early embryonic condition. The origin of adrenal cortical tissue and mesonephric tissue from the same source is unmistakable in the normal development of *Amblystoma punctatum*.

These observations agree with the reports of neither Albrand (1908) on *Amblystoma tigrinum* nor Stenger and Charipper (1946) on *Rana pipiens*. They have described the adrenal cortical anlagen as originating from the coelomic epithelium. Crowder (1957) described the origin of human adrenal cortical cells from coelomic and mesonephric epithelium. The observations in this investigation are in agreement with the reports of Segal (1953) and Chang and Witschi (1955), who claim a mesonephric anlagen origin for the frog adrenal cortical tissue.

MIGRATION PATHWAYS AND ADJACENT TISSUE. The classic experiments of Holtfreter (1939, 1947) clearly show that the adhesiveness between cell membranes is an important factor in cell migration. For that reason, the sequence of contact of cortical cells with various tissues was noted.

It was observed that adrenal cortical lobules first adhered to the vena cava. They appeared to migrate from the dorsal side along the lateral walls to the ventral side of the vena cava during the course of larval life. This agrees with observations of Branin (1937) and Segal (1953). New mesonephric tubules first appeared dorsal to the adrenal cortical tissue, and in their growth they appeared to force the cortical tissue into more ventral levels. Some adrenal cortical lobules appeared to be pushed laterally along the efferent renal veins. It was not possible to determine how much, if any, migration was intrinsic to the cortical cells. It is signficant that all adrenal cortical tissue in *Amblystoma punctatum* adults was seen adhering to the vena cava or to the blood sinuses of efferent renal veins (Figs. 1 and 4).

The above observations are in disagreement with observations on *Triton alpestris* and *Triton cristatus* by Gray (1932), who described migration of adrenal cortical cells primarily along mesonephric tubules. The present investigation confirms graft experiments by Williams (1954), who found that adrenal cortical grafts aligned along veins of the host. They also agree with Branin's (1937) notation of the migration of adrenal cortical cells along the vena cava.

Detwiler (1937) described the early migration of sympathoblasts derived from the vitally stained neural crest of *Amblystoma punctatum* before feeding. He reported their migration between the myotomes and the neural tube

and notochord to a location considerably ventral to the region where spinal ganglia were to form. They were among the first cells to migrate from the neural crest, and he suggested that they might be the developing sympathetic ganglia. It is generally accepted that some sympathoblasts migrate from the developing sympathetic ganglia to more ventral locations, where they become adrenal medullary cells or chromaffin body cells. (Refer to reviews by Hollinshead, 1940; van Campenhout, 1930a; Yntema and Hammond, 1947.)

In the present investigation at the beginning of feeding (10–12 mm.) sympathoblasts were observed migrating along the notochord toward the developing sympathetic ganglia. Spinal ganglia were forming at this time. A few days later (11–15 mm.) sympathoblasts were seen adhering to the latero-ventral surface of the aorta. Some were close to the developing adrenal cortical anlagen (Fig. 3). Evidence from a study of later stages indicates that these sympathoblasts were probably the precursors of both the adrenal medullary cells and the cells of the chromaffin bodies. As the vena cava expanded dorsally, sympathoblasts were seen adhering to it. In the same sections sympathoblasts were seen in contact with adrenal cortical cells. During late larval and adult life many cases were observed of anterior cortical lobules which completely surrounded the medullary cells (Fig. 5). In posterior adrenal cortical lobules, adrenal medullary cells were adjacent to the lobules. Some posterior lobules lacked adrenal medullary cells (Fig. 1).

DIFFERENTIATION RELATIONS BETWEEN ADRENAL COMPONENTS. Several investigators of adrenal differentiation have suggested an inductive sequence involving the two adrenal components. It has been demonstrated that cortical-cell differentiation is independent of adrenal medullary cells (van Campenhout, 1930b; Hammond and Yntema, 1947). The observations in the present investigation confirmed these conclusions.

The differentiation of adrenal medullary cells has been described by Kuntz (1912) and van Campenhout (1930b) as being dependent upon the direct diffusion of hormones secreted by the adrenal cortical cells to adjacent sympathoblasts. This causal relationship may be necessary; however, a careful study of serial sections in the present investigation showed many cases of chromaffin cells separated from adrenal cortical cells by as many as four kidney tubules in larvae as well as in adults. In Figures 1 and 4 it can be seen that the common location factor for chromaffin adrenal cells and cells of chromaffin bodies is in juxtaposition along the vena cava or efferent renal veins, not in contact with adrenal cortical cells.

The pseudo-segmental appearance of medullary cells and of chromaffin bodies can be clearly seen (Fig. 1). The pseudo-segmental arrangement of all chromaffin tissue along the vena cava occurs whether or not cortical cells are present.

DIFFERENTIATION OF CHROMAFFIN CELLS. Histological differentiation of chromaffin cells was observed to begin during mid-larval life. At that time the maturation process was visible in changes in cell size and in the appearance of chromaffin granules. In larvae of 25 mm. (total length) the chromaffin reaction was observed in cells that bordered the vena cava whether or not they were in contact with adrenal cortical tissue. The simultaneous and similar differentiation of both adrenal medullary cells and chromaffin body cells is the primary basis for assuming the equivalence of these two morphological categories of chromaffin bodies. At that time they both develop a

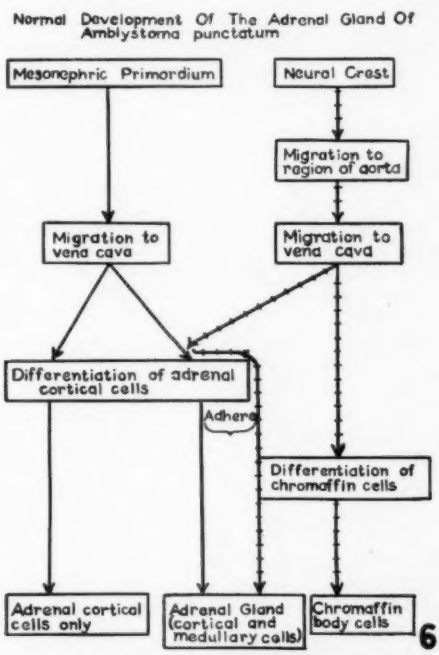


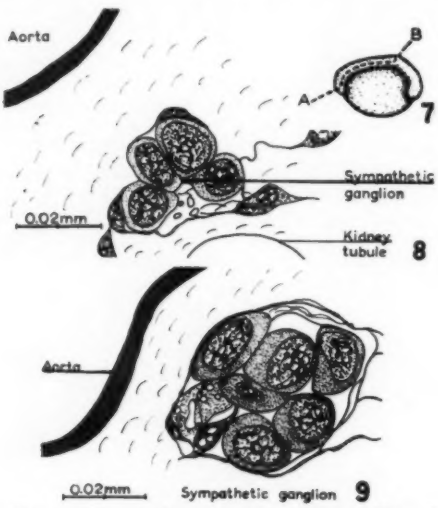
FIG. 6. Diagram summarizing normal development of adrenal cortical and chromaffin tissue in *Amblystoma punctatum* indicating chronology of migration, contact with other tissues, and differentiation.

distinctive glandular appearance in contrast to other sympathoblasts which develop typical nerve-cell processes. The similarity in adrenal medullary cells and the chromaffin body cells can be seen in Figure 2.

SUMMARY OF NORMAL DEVELOPMENT. Figure 6 summarizes the observations of normal development in *Amblystoma punctatum*.

Experiments on the Bilateral Extirpation of Trunk Neural Crest

The trunk neural folds were removed from 100 *Amblystoma punctatum* neurulae at stage 18–19 to determine the effects of extirpation on the development of adrenal components. Figure 7 illustrates the manner in which the operations were performed. Special precautions were taken to remove only the neural folds as indicated. Deformed embryos were discarded since deformity might affect the normal formation of somites and the segmentation of nervous tissue known to be dependent upon the metamerism of the myotomes (Lehmann, 1927; Detwiler, 1934, 1935). Embryos which showed evidence that the operation had not removed all trunk neural crest were also discarded. External criteria for an unsuccessful operation were the presence of any dorsal fin or pigment cells in the trunk region. Embryos free of trunk neural crest along with their unoperated controls were killed and fixed *in toto* from stage 46 (9 mm.) to premetamorphic stages (42 mm). Twenty-six crest-free animals were used in the histological study. Serial cross sections were made of the entire body at the level of the mesonephroi. The relative number of sympathetic ganglion cells and adrenal medullary cells was obtained by counting these cells in every third section of a 200-section ribbon taken from each experimental and control animal.



FIGS. 7–9. *Amblystoma punctatum*. FIG. 7. Neurula showing location of neural fold excision between A and B. FIG. 8. Largest sympathetic ganglion in a 42-mm. larva devoid of trunk neural crest. Note lack of connective tissue sheath and smaller size than control. FIG. 9. Sympathetic ganglion in a control 45-mm. larva.

RESULTS. Spinal ganglia were missing in all animals devoid of trunk neural crest. Abortive sympathetic ganglia were found in all trunk crest-free larvae above 29-mm. total body length. The largest ganglion found is shown in Figure 8. The cells are normal in appearance but reduced in numbers (Figs. 8 and 9). Abortive ganglia contained from one to four cells. Controls of comparable size contained from four to ten cells per sympathetic ganglion. Nine-tenths of the abortive ganglia lacked connective tissue sheaths and looked like the sympathetic ganglia in young control larvae (Fig. 3). The reduction in total number of sympathetic ganglion cells is indicated in Table I.

Table I

The relative number of sympathetic ganglion cells and adrenal medullary cells in crestless and normal larvae.

Size of larvae (mm.)	Sympathetic Ganglia Cells		Adrenal Medullary Cells	
	Crest-less	Average No. in Controls	Crest-less	Average No. in Controls
15	0	31	0	0
20	0	88	0	10
25	0	—	0	—
30	6	100	0	22
31	7	—	1	—
33	20	—	1	—
33	23	—	2	—
38	27	102	19	76
42	35	160	12	96

It can be seen that the sympathetic ganglion cells appeared prior to the adrenal medullary cells in the experimental animals and that the latter cells were also reduced in numbers. In the largest crestless larvae (42 mm.) there were 22 per cent of the normal complement of sympathetic ganglion cells and 12 per cent

of the adrenal medullary cells found in controls of comparable size.

SUMMARY. Adrenal medullary cells and sympathetic ganglion cells can form in later larval life in animals devoid of trunk neural crest. The observations above do not concur with the report of van Campenhout (1930b) that removal of trunk neural crest in *Rana pipiens* and *Rana palustris* prevented the formation of adrenal medullary cells and sympathetic ganglia. It appears that van Campenhout (1930b) did not carry his crestless animals (maximum 20 mm.) long enough to observe the late appearance of sympathetic ganglia and adrenal medullary cells. These results agree with the findings of D. S. Jones (1939), who reported sympathetic ganglia in crestless chicks in late embryonic life. Speculations on the possible origin of these medullary cells will be outlined in the discussion.

EFFECT OF EXTIRPATION OF NEURAL CREST ON ADRENAL CORTICAL CELLS. Van Campenhout (1930b) reported that in spite of the fact that it is of mesodermal origin, there is a decrease in the normal amount of adrenal cortical tissue in animals devoid of trunk neural crest. He maintained the frogs devoid of trunk neural crest to only 20 mm. and did not specify the number of cortical cells or any change in their numbers. Branin (1937) established that in salamanders, proliferation of adrenal cortical

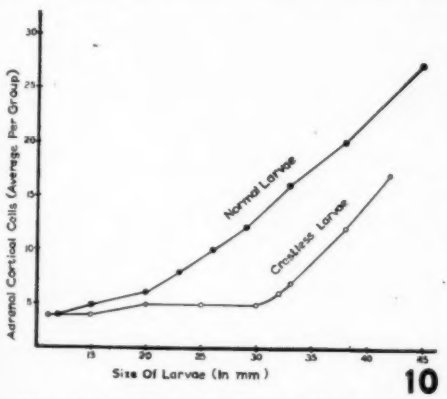


FIG. 10. Graph showing the relative number of adrenal cortical cells in *Amblystoma punctatum* larvae devoid of trunk neural crest and in control larvae of designated sizes.

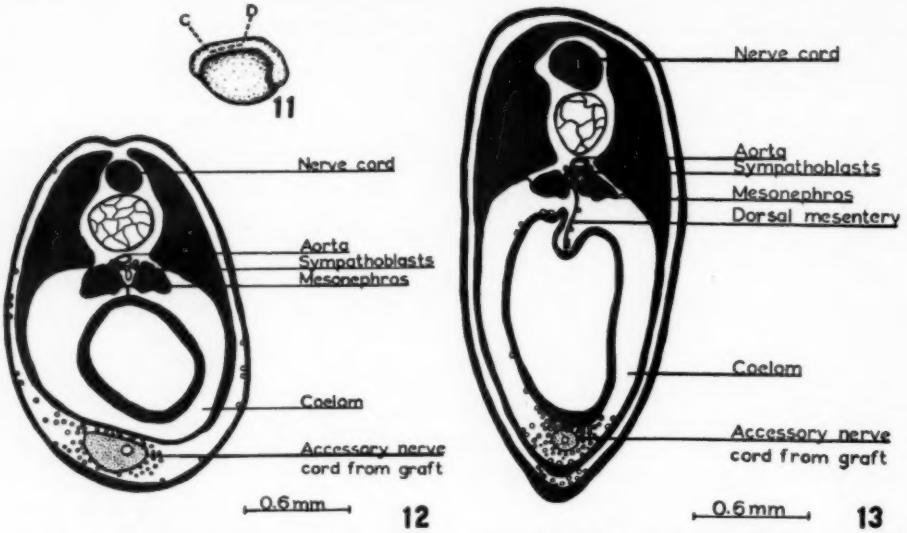
cells takes place throughout larval life, increasing both the number of cortical islands and the number of cells in each island group. In the present investigation a count was made of the number of cells per group in every third section in the mesonephric region of both experimental animals and of controls. From these figures the average number of cells per cortical group was calculated for each stage. These averages represent relative numbers and not absolute numbers of cells. The results of these calculations are plotted graphically in Figure 10 and provide a quantitative basis for comparing the number of adrenal cortical cells in control and crest-free embryos. This graph demonstrates that after an initial lag extending to the 30-mm. stage, the number of adrenal cortical cells in the experimental larvae increased at a rate comparable to that observed in controls.

SUMMARY. The above observations show that secondary proliferation of adrenal cortical cells can take place in the absence of trunk neural crest and that whatever may be the early dependence or stimulus of cortical cell proliferation by medullary cells is of temporary duration. The present observations indicate that the cortical cells can multiply in a normal manner in the absence of all but a very small percentage of the normal medullary component of the adrenal complex (Table I).

Transplantation of Neural Crest Mid-Ventrally on Embryos Deprived of Trunk Neural Crest

The neural crest and its diverse fates have been the subject of many embryological investigations in recent years. A great body of information has been accumulated on certain neural crest derivatives (*i.e.*, visceral cartilages, pigment cells, etc.), but relatively little is known about the medullary cells of the adrenal complex and the chromaffin bodies (Du Shane, 1943, 1944; Rawles, 1948; Hörstadius, 1950). For example, whether or not the adrenal medullary cells in *Amblystoma punctatum* are rigidly determined when they begin their migration from the neural crest has not been established. It has been demonstrated that the differentiation of pigment cells and preganglion cells from the neural crest is greatly influenced by extrinsic factors in their normal tissue environments (reviews by Hörstadius, 1950; Lehman, 1950, 1951). It seemed appropriate to analyze the development of the adrenal medullary cells of *Amblystoma punctatum* under conditions which altered the tissue environments with which they are normally associated. To this end a series of experiments involving two operations was made.

Crestless embryos were prepared by making bilateral extirpations of trunk neural folds as



FIGS. 11–13. *Amblystoma punctatum.* FIG. 11. Neurula showing location of excision between C and D, from which donor graft was taken. FIG. 12–13. Transverse section of 15-mm. crestless host larvae each with neural crest graft. Cells from graft are indicated as small circles and include a composite record of locations of donor cells over anterior-posterior extent of graft. Note sympathoblasts between aorta and vena cava. FIG 12. Graft in body wall. FIG. 13. Graft in coelomic cavity.

described above. This was followed by a second operation involving the grafting of stage 18–19 donor trunk neural folds onto the crestless host embryos at stage 35 (Figs. 7 and 11). Based on the criteria described above, only embryos completely free of trunk neural crest were used for the ventral graft experiments. Grafts were held in place with glass bridges until they healed into place. The animals were fixed *in toto* two weeks after feeding had begun, when they were approximately 15 mm. in length. Ten animals with successful ventral neural crest grafts located at the level of the mesonephroi were selected for histological study. Migration pathways and differentiation of neural crest cells were recorded from cross sections at the level of the graft in composite camera lucida drawings covering the anterior-posterior extent of the graft (Figs. 12, 13).

RESULTS. Two to four days after the grafts were made, pigment cells appeared first around the graft and later on the dorsal side of the animal opposite the graft. Since no pigment cells occur on the trunk in trunk-crest-free animals at this stage, the pigment cells described were interpreted as migrating from the grafted neural crest.

Table II shows the relative number of sympathoblasts found adjacent to the aorta in seventy-five consecutive cross sections at the level of the mesonephroi. When one compares the number of sympathoblasts in crestless hosts bearing ventral grafts of neural crest (column 1) with control larvae of the same age (column 2), he can readily see that the experimental larvae have somewhat fewer sympathoblasts than the controls. However, the comparison

Table II

The relative number of sympathoblasts in crestless host larvae with neural crest grafts, control larvae, and crestless larvae.

Case Number	Sympathoblasts in Larvae with Ventral Grafts	Sympathoblasts in Control Larvae	Sympathoblasts in Crestless Larvae
EV71	10	14	0
EV73	8	13	0
EV74	11	14	0
EV75	8	12	0
EV76	13	11	0
EV 8	8	12	0
EV 9	12	15	0
Totals.....	70	91	0

between the first and third columns is much more striking and is taken as conclusive evidence that the sympathoblasts found in experimental embryos with ventral grafts came from the graft. Since the presence of the ventral graft is the only way in which these embryos (column 1) differed from crestless embryos (column 3), there seems little doubt that the grafted crest cells have migrated around the coelomic wall to the normal site for sympathetic ganglion formation.

The evidence indicates that migration pathways are not a significant factor in the differentiation of sympathoblasts. When the graft was located entirely within the body wall, the majority of the cells adhered to the epidermis and traveled as far dorsally as the nerve cord. In these cases sympathoblasts were observed between the aorta and vena cava and along the mesial edges of the mesonephroi in the typical location for sympathoblasts in the controls (Fig. 12). When the graft was located in the coelomic cavity, neural crest cells were observed along the ventral and dorsal mesentery and in the mesonephroi around the aorta and vena cava (Fig. 13). Thus the sympathoblasts are in the positions normally occupied by the sympathetic ganglia and adrenal medullary cells. Note also the absence of spinal ganglia, none of which were observed in the crestless larvae although they are present in the controls at this stage.

SUMMARY. The above data give evidence that sympathoblasts are able to develop from neural crest grafts placed ventrally as described. Since it has been shown that some sympathoblasts in this area give rise to adrenal medullary cells, the conclusion is that the pathway to the vena cava taken by migrating neural crest is not important to the determination of sympathoblasts nor to adrenal medullary cells. It is then not necessary that the potential adrenal medullary cells migrate from the neural crest ventrally between the somites and neural tube nor by the notochord as they do in normal development.

Discussion

Morphogenesis of the Adrenal Glands of *Amblystoma punctatum*

From the study of normal development in *Amblystoma punctatum* certain conclusions can be drawn which are substantiated by the experimental results. In the development of many organs (*i.e.*, eye, ear) whose components originate from different germ layers, an inductive sequence involving contact of the components can be demonstrated. The data in the present investigation is interpreted as evidence that contact between the adrenal cortical and adrenal medullary cells is unnecessary for the differentiation, migration, and proliferation of either component.

DIFFERENTIATION. Differentiation of adrenal cortical cells occurred without contact with the adrenal medullary cells. This supports the findings on anura by van Campenhout (1930b) and Stenger and Charipper (1946).

Differentiation of chromaffin cells was shown to occur without contact from the adrenal cortical cells (Figs. 1, 3, and 6). Kuntz (1912) and van Campenhout (1930b) suggested that the differentiation of adrenal medullary cells is dependent upon hormones from the adrenal cortical cells secreted directly into the surrounding tissue. C. I. Jones (1957) and Liu and Maneely (1959) pointed out that the methylation of noradrenalin to adrenaline, which depends on cortical secretion, might be related to the central adrenal medullary tissue. The position of both adrenal components along common blood sinuses in the present investigation has been interpreted as evidence that diffusion from one adrenal cell type to the other is possible, but that contact of cortical and medullary cells is not necessary for their differentiation.

MIGRATION. The migration of future adrenal medullary cells to the adrenal cortical cells was described by Kuntz (1912) as stimulated and tropistically directed by the adrenal cortical hormones, which form a diffusion gradient in the surrounding tissue. It seems highly improbable that Kuntz's explanation is adequate to account for the great distances traveled by the future adrenal medullary cells, both in normal development and in the experiments of grafted neural crest.

The intrinsic capacity to migrate has been demonstrated for many neural crest cells Detwiler, 1937; Twitty and Niu, 1948). In the present investigation the migratory ability of future sympathoblasts was observed in the ventral graft experiments (Figs. 12 and 13). That available space and the mechanical factors involved in growth can account for some morphological movements is known. These mechanical factors alone appear inadequate to explain the direction of the pathways through mesenchyme taken in normal development and in the ventral graft experiments.

Holtfreter's classic experiments (1939, 1947) demonstrated the importance of the cohesive-

ness between cell membranes in cell migration and organogenesis. The observations on normal development of the adrenal show a chronological sequence of contact of the adrenal components with specific adjacent tissue—namely, the vena cava. The observations also show that the adrenal components adhere more strongly to certain tissues than to other tissues. On the basis of Holtfreter's experiments (1939, 1947) and the results of the present investigation,

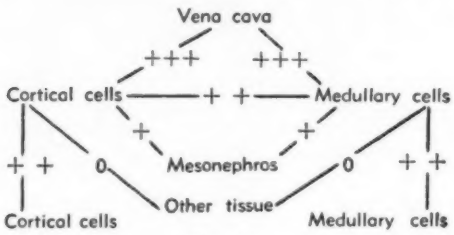


FIG. 14.

the tissue affinities shown in Figure 14 for the developing adrenal components are postulated in terms of relative intensity (0 to +++).

It can be seen that the common location factor for adrenal cortical cells, adrenal medullary cells, and chromaffin body cells is the vena cava and its branches (Figs. 1 and 4). Differentiation of these cells takes place after this contact has been made. The importance of the vena cava in the morphogenesis of the adrenal glands of amphibia is further illustrated by the results of the ventral graft experiments. The fact that the sympathoblasts appeared only at the vena cava is interpreted as evidence that the capacity for self-differentiation as chromaffin cells cannot be expressed unless they enter the normal terrain for adrenal medullary-chromaffin differentiation. The normal terrain for the adrenal components is interpreted as the region of the vena cava.

PROLIFERATION. Van Campenhout (1930b) and Hammond and Yntema (1947) described a decrease in the normal complement of adrenal cortex of trunk neural crest-free embryos. In the present investigation larvae devoid of trunk neural crest showed an increase in secondary adrenal cortical proliferation which closely paralleled the normal proliferation rate (Fig. 10). This indicates that cell multiplication in a normal manner can take place in the absence of all but a relatively small percentage of the normal complement of the adrenal medulla.

Data from the present investigation indicate an indirect effect of adrenal components upon one another. For example, Amblystoma larvae devoid of trunk neural crest never contained as much adrenal cortex as normal larvae. Segal (1953) and Chang and Witschi (1955) found hyperplasia in both adrenal components when they raised frog larvae in estradiol solutions. Both reports suggested that the hyperplasia was probably mediated through an undetermined hormonal mechanism involving the anterior pituitary. It is possible that a related mechanism might be involved in the decreased stimulus to adrenal cortical proliferation noted above.

Regulation in Adrenal Medullary Cells and Sympathetic Ganglia

In the present investigation the late appearance of adrenal medullary cells and of sympathetic ganglia in Amblystoma larvae (from 28 mm. to 42 mm.) devoid of trunk neural crest is interpreted as a case of regulation in which the embryo compensates for experimental loss of a normal primordium by recruiting replacement cells from another source. The absence of these cells in the young crest-free larvae in the present study and in the observations of van Campenhout (1930b) indicates that trunk neural crest normally contributes extensively during early larval life to the adrenal medulla and sympathetic ganglia. It appears that van Campenhout (1930b) did not carry his crestless animals (maximum length 20 mm.) long enough to observe the late appearance of these cells.

The sympathoblasts normally appear at the adrenal cortical cells in an anterior-posterior time sequence. The adrenal medullary cells observed in crestless Amblystoma larvae from 28 mm. to 42 mm. were at middle levels of the mesonephroi and were not at the anterior or posterior extremities. If the cells were coming from a source which normally contributes sympathoblasts to the adrenal medulla, one would expect the same anterior-posterior sequence of their appearance. The fact that adrenal medullary cells appeared only in the center of the mesonephroi is interpreted as evidence of a source of cells which normally do not contribute or do not contribute extensively to the adrenal medullary complement. It is possible that the adrenal medullary deficiency in crestless larvae stimulates the conversion of labile cells (which would normally form something else) to become adrenal medullary cells. The present investigation does not give evidence for the source of the labile sympathoblasts in animals devoid of trunk neural crest. From investigations of the origin of sympathetic ganglia, the following possibilities are suggested:

1. Migration from neural crest of the head or tail.
2. Origin from a nonerest source such as the ventral part of the nerve cord.

Many investigators have contributed data which give evidence of a neural crest origin for the sympathoblasts and their cell progeny (van Campenhout, 1930a, 1930b; Detwiler, 1937; Yntema and Hammond, 1947; Hammond and Yntema, 1947). Anterior-posterior migration of neural crest has been demonstrated (Detwiler, 1937; Du Shane, 1943; Twitty, 1949), and it is possible that these cells might have migrated from the head or tail neural crest.

Other sources for the sympathoblasts have been suggested. Raven (1937) concluded from experiments on xenoplastic grafts of neural crest and ventral neural tube from *Amblystoma punctatum* to *Triturus taeniatus* that sympathetic ganglia in urodeles come from the ventral part of the nerve cord as well as from the neural crest. D. S. Jones (1937, 1939, 1941) confirmed the above results when he excised the ventral part of the nerve cord of the chick and found a lack of sympathetic ganglia. It is possible that the adrenal medullary cells and sympathetic ganglia in crestless larvae of the present study might have originated from sympathoblasts from the ventral part of the nerve cord. This investigation revealed no negative or positive evidence for either possibility. Since there was very slight regulation of adrenal medullary cells, this study concludes that most of the adrenal medulla normally is derived from trunk neural crest and is not completely replaced by other primordial cell types.

Phyletic Implications

Between elasmobranchs and mammals phyletic differences in the relative position and amounts of the two components exist, but deviations from the normal picture for the class type have been found (Fraser, 1930, in sharks; Segal, 1953, and Chang and Witschi, 1955, in frogs; Nelson, 1939; Lasher, 1947; Graham, 1953, in humans). These reports and the present investigation indicate that the factors involved in the morphogenesis of the vertebrate adrenals are dependent upon similar factors.

Class differences in adrenals might then be explained in terms of differences in amounts of mesonephric blastema, intensities of stimuli, and differences in timing. In amphibia the mesonephric primordia extends over a greater part of the coelomic cavity. The mesonephric primordia normally give rise to scattered adrenal cortical islands the length of the mesonephros. Contact between chromaffin cells and adrenal cortex is insured only if the sympathoblasts reach the region of the vena cava at a time when and at a place where an adrenal cortical lobule has formed. If this does occur, the two adrenal components adhere; if it does not occur, the adrenal components remain separated but adhere to the vena cava (Fig. 6). Normally, as the mesonephroi are beginning to form, the intensity of the stimulus is greater for the formation of adrenal cortex than for the formation of the other derivatives of the mesonephric primordium (nephrons and gonad medulla, Witschi, 1951a, 1951b). The migrating sympathoblasts in this area are then more likely to make contact with adrenal cortical lobules between which they continue their migration for a short time. The cohesive force between the adrenal components is great. Later, when secondary proliferation of cortex occurs, the cortical cells proliferate around the adrenal medullary cells. Thus there were some anterior lobules found in which the adrenal cortical cells completely surrounded the adrenal medullary cells. In posterior lobules which formed later, the adrenal cortical lobule formation does not coincide with sympathoblast migration. As a consequence, the adrenal components are separated at posterior levels.

Since the normal urodele adrenal resembles the anomalous condition reported in man, it is suggested that the present investigation has medical implications. The critical period for the formation of accessory adrenal cortical nodules apparently occurs between three and nine weeks, when the mesonephric blastema may be induced to form adrenal cortical cells. Depending on the relative timing of sympathoblast stimulation and adrenal-cortical-cell induction, the various combinations of accessory adrenals would result.

Summary

The morphogenesis of adrenal glands of *Amblystoma punctatum* was observed after two series of operations. The first involved the bilateral extirpation of trunk neural crest. The second series included the grafting of trunk neural crest mid-ventrally on embryos previously deprived of their own trunk neural crest. For comparison, normal development was observed of both adrenal components from stage 46 to the 175-mm. adult. This investigation focused on the interrelation of the adrenal components.

Although the trunk neural crest has been considered by some investigators, the source of adrenal medullary cells and sympathetic ganglia, in the salamanders devoid of trunk neural crest, 12 per cent of the normal adrenal medullary cell complement and 22 per cent of the normal number of sympathetic ganglia did appear in late larval life. This is in disagreement with the findings of van Campenhout (1930b) in the frog. The conclusion is that although most of the adrenal medulla in urodeles normally is derived from trunk neural crest, it is only partially replaceable by other primordial cell types.

Adrenal cortical cells were less numerous in larvae devoid of trunk neural crest than in controls. A sharp increase in number of adrenal cortical cells occurred after 30 mm., when sympathoblasts appeared. It is suggested that secondary proliferation of adrenal cortical cells is probably mediated through a hormonal mechanism involving the anterior pituitary.

Cells from neural crest homografts placed in a ventral position in *Amblystoma punctatum* larvae devoid of trunk neural crest migrated to the region of the vena cava, where they became sympathoblasts. The evidence indicates that normal pathways of migration are not essential for the development of sympathoblasts. The capacity for self-differentiation as chromaffin cells was not expressed unless the sympathoblasts entered the normal terrain for adrenal medullary cells (the area of the vena cava).

An inductive sequence involving contact between adrenal components was found not essential for their differentiation and proliferation. The importance of the vena cava in the morphogenesis of amphibian adrenals was demonstrated. The conclusion is that the following factors are involved in the morphogenesis of adrenal glands: intrinsic migratory powers of the neural crest, tissue affinities, differentiation of mesonephric blastema, hormonal stimulation, and relative timing of migration of adrenal medullary cell primordia and differentiation of cortical cells.

An explanation for the development of different types of accessory adrenals in man was discussed in the light of the findings presented here of a similar type of adrenal complex found normally in urodeles.

LITERATURE CITED

ALBRAND, M. 1908. Die Anlage der Zwischenniere bei den Urodelen. Arch. f. mikr. Anat. **72**: 353–385.

BRANIN, M. L. 1937. The development of the cortical adrenal in the four-toed salamander *Hemidactylium scutatum* (Schlegel). J. Morph. **60**: 521–561.

CHANG, C. Y., AND E. WITSCHI. 1955. Independence of adrenal hyperplasia and gonadal masculinization in the experimental adrenogenital syndrome of frogs. Endocrin. **56**: 597–605.

CROWDER, R. E. 1957. The development of the adrenal gland in man, with special reference to origin and ultimate location of cell types and evidence in favor of the "Cell Migration Theory." Contributions to Embryology **36**: 242–251. Carnegie Institute of Washington. Pub. 611. Washington, D. C.

DETWILER, S. R. 1934. An experimental study of spinal nerve segmentation in Amblystoma with reference to the plurisegmental contribution to the brachial plexus. J. Exp. Zool. **67**: 395–441.

———. 1935. The development of spinal ganglia following transplantation of the spinal cord with or without somites. Anat. Rec. **61**: 441–455.

———. 1937. Observations upon the migration of neural crest cells and upon the development of the spinal ganglia and vertebral arches in amblystoma. Am. J. Anat. **61**: 63–94.

DU SHANE, G. P. 1943. The embryology of vertebrate pigment cells. Part I. Amphibia. Quart. Rev. Biol. **18**: 109–127.

———. 1944. The embryology of vertebrate pigment cells. Part II. Birds. Quart. Rev. Biol. **19**: 98–117.

FRASER, A. H. H. 1930. Lipin secretion in the elasmobranch inter-renal. J. Micr. Soc. **73**: 121–134.

GRAHAM, L. S. 1953. Celiac accessory adrenal glands. Cancer **6**: 149–152.

GRAY, P. 1932. The development of the amphibian kidney. Quart. J. Micr. Sci. **75**: 425–465.

HAMMOND, W. S., AND C. L. YNTEMA. 1947. Depletions in the thoracolumbar sympathetic system following removal of neural crest in the chick. J. Comp. Neur. **86**: 237–267.

HOLLINSHEAD, W. H. 1940. Chromaffin tissue and paraganglia. Quart. Rev. Biol. **15**: 156–171.

HOLTFRETER, J. 1939. Gewebeaffinität, ein Mittel der embryonalen Formbildung. Arch. f. exp. Zellforsch. **23**: 169–209.

———. 1947. Observations on the migration, aggregation and phagocytosis of embryonic cells. J. Morph. **80**: 25–55.

HÖRSTADIUS, S. 1950. The Neural Crest, Its Properties and Derivatives in the Light of Experimental Research. Oxford University Press: London.

JONES, C. I. 1957. The Adrenal Cortex. Cambridge University Press: London.

JONES, D. S. 1937. The origin of the sympathetic trunks in the chick embryo. Anat. Rec. **70**: 45–62.

———. 1939. Studies on the origin of sheath cells and sympathetic ganglia in the chick. Anat. Rec. **73**: 343–351.

———. 1941. Further studies on the origin of sympathetic ganglia in the chick embryo. Anat. Rec. **79**: 7–13.

KUNTZ, A. 1912. The development of the adrenals in the turtle. Am. J. Anat. **13**: 71–88.

LASHER, E. P. 1947. Primary extrarenal hypernephroma; discussion of sites and origins, and report of a case. West. J. Surg., **55**: 87–93.

LEHMAN, H. E. 1950. The suppression of melanophore differentiation in salamander larvae following orthotopic exchange of neural folds between species of Amblystoma and Triturus. J. Exp. Zool. **114**: 435–464.

———. 1951. An analysis of the dynamic factors responsible for the phenomenon of pigment suppression in salamander larvae. Biol. Bull. **100**: 127–152.

LEHMANN, F. E. 1927. Further studies on the morphogenetic role of somites in the development of the nervous system of amphibians. The differentiation and arrangement of the spinal ganglia in *Pleurodeles waltli*. J. Exp. Zool. **49**: 93–132.

LIU, H. C., AND R. B. MANEELY. 1959. Some aspects of adrenal histology and histochemistry in the soft-shelled turtle of South China (*Chelonia amyda*). J. Endocrin. **18**: 1–7.

NELSON, A. A. 1939. Accessory adrenal cortical tissue. Arch. Path. **27**: 955–965.

RAVEN, C. P. 1937. Experiments on the origin of the sheath cells and sympathetic neuroblasts in amphibia. J. Comp. Neur., **67**: 221–240.

RAWLES, M. E. 1948. Origin of melanophores and their role in development of color patterns in vertebrates. Physiol. Rev. **28**: 383–408.

SEGAL, S. J. 1953. Morphogenesis of the estrogen induced hyperplasia of the adrenals in larval frogs. Anat. Rec. **115**: 205–230.

STENGER, A. H., AND H. A. CHARIPPER. 1946. A study of adrenal cortical tissue in *Rana pipiens* with special reference to metamorphosis. J. Morph. **78**: 27–40.

TWITTY, V. C. 1949. Developmental analysis of amphibian pigmentation. Growth Symp. **9**: 133–161.

TWITTY, V. C., AND M. C. NIU. 1948. Causal analysis of chromatophore migration. J. Exp. Zool. **108**: 405–438.

VAN CAMPENHOUT, E. 1930a. Historical survey of the development of the sympathetic nervous system. Quart. Rev. Biol. **5**: 217–234.

———. 1930b. Contribution to the problem of the development of the sympathetic nervous system. J. Exp. Zool. **56**: 295–320.

WILLIAMS, R. G. 1954. Microscopic studies in living mammals with transparent chamber methods. In International Review of Cytology, ed. G. H. Bourne and J. F. Danielli. III: 382–383.

WITSCHI, E. 1951a. Embryogenesis of the adrenal and the reproductive glands. Recent Progr. Hormone Res. **6**: 1–27.

———. 1951b. Adrenal hyperplasia in larval frogs treated with natural estrogens. Anat. Rec. **111**: 35–36.

YNTEMA, C. L., AND W. S. HAMMOND. 1947. The development of the autonomic nervous system. Biol. Rev. **22**: 344–359.

CYCLICLY RELATED DIFFERENTIAL EQUATIONS AND THE CIRCULANT[1]

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Introduction

W. M. Whyburn [2] was led from a consideration of simple systems of ordinary differential equations to consider the nature of the solutions of

$$(1)\quad y_i' = \sum_{k=1}^{n} A_k\, y_{i+m+hk}, \qquad (i = 1, \ldots, n;\ y_{j+n} \equiv y_j),$$

where h and m are integers and the coefficients A_k are integrable functions on the closed interval $[a, b]$. He exhibited the exponential nature of the solutions of (1) by obtaining directly, with the aid of the Lagrange resolvent and its properties, the general solution of the equations. The present paper uses matrix methods, in particular the theory of the circulant, to solve (1). It includes a treatment of

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$$y_i' = \sum_k A_k\, y_{i+a(k)}, \quad (i=1, \ldots, n;\ y_{j+n} \equiv y_j),$$

and

$$y_i' = \sum_{k,l} A_{kl}\, y_{i+a_l(k)}, \quad (i=1, \ldots, n;\ y_{j+n} \equiv y_j).$$

The Circulant

Let $a_0, a_1, \ldots, a_{n-1}$ denote functions defined on a common domain and whose range of values are subsets of the real or complex field. The n-square matrix $A = (a_{ij})$ is called a circulant (cf. [1], Chapter VII, section 29) whenever

$$a_{ij} = \begin{cases} a_{j-i}, & j \geqq i \\ a_{j-i+n}, & j < i \end{cases} \tag{2}$$

so that

$$A = \begin{bmatrix} a_0 & a_1 & a_2 & \ldots & a_{n-2} & a_{n-1} \\ a_{n-1} & a_0 & a_1 & \ldots & a_{n-3} & a_{n-2} \\ a_{n-2} & a_{n-1} & a_0 & \ldots & a_{n-4} & a_{n-3} \\ \cdot & \cdot & \cdot & \cdot & \cdot & \cdot \\ a_1 & a_2 & a_3 & \ldots & a_{n-1} & a_0 \end{bmatrix}.$$

Note that the circulant is symmetric with respect to the diagonal through the a_{1n} and a_{n1} elements, and also that the transpose, the conjugate, and the conjugate transpose of a circulant are also circulants.

Theorem 1. There exists a non-singular matrix T such that, for every n-rowed circulant A, $AT = TD$, where D is a diagonal matrix.

Proof. Let $r_1, \ldots, r_n$ be the n distinct n-th roots of unity described by

$$r_k = \cos \frac{2k\pi}{n} + i \sin \frac{2k\pi}{n}.$$

Take

$$T = (t_{ij}) = (r_j^i) = \begin{bmatrix} r_1 & r_2 & \ldots & r_n \\ r_1^2 & r_2^2 & \ldots & r_n^2 \\ \cdot & \cdot & \ldots & \cdot \\ r_1^n & r_2^n & \ldots & r_n^n \end{bmatrix}. \tag{3}$$

Since $r_j^i = r_i^j$, one has $T = (r_j^i) = (r_i^j)$; that is, T is a symmetric matrix. That T is non-singular follows from the fact that (r_i^j) is equivalent to a Vandermonde matrix and that every Vandermonde matrix is non-singular. Now consider $AT = (c_{ij})$. In view of (2),

$$\begin{aligned} c_{ij} &= \sum_{k=1}^{n} a_{ik}\, r_j^k \\ &= a_{ii}\, r_j^i + \Big(\sum_{k=1}^{i-1} + \sum_{k=i+1}^{n}\Big) a_{ik}\, r_j^k \\ &= r_j^i \Big(a_0 + \sum_{k=1}^{i-1} a_{k-i+n}\, r_j^{k-i+n} + \sum_{k=i+1}^{n} a_{k-i}\, r_j^{k-i}\Big). \end{aligned}$$

The first and second sums can be rewritten as

$$\sum_{k=n-i+1}^{n-1} a_k\, r_j^k \text{ and } \sum_{k=1}^{n-i} a_k\, r_j^k,$$

respectively. Thus,

$$c_{ij} = r_j^i \sum_{k=0}^{n-1} a_k\, r_j^k.$$

Now set $D = (d_{ij})$ with

$$d_{ij} = \begin{cases} \sum_{k=0}^{n-1} a_k\, r_j^k, & i = j \\ 0, & i \neq j \end{cases} \tag{4}$$

It follows that $TD = (c_{ij})$. For,

$$\sum_{k=1}^{n} t_{ik}\, d_{kj} = t_{ij}\, d_{jj} = r_j^i\, d_{jj} = c_{ij}.$$

Cyclicly Related Equations

Let $A_1(x), \ldots, A_n(x)$ be real functions of the real variable x defined on the bounded closed interval $[a, b]$. Consider the system of differential equations

$$y_i' = \sum_{k=1}^{n} A_k\, y_{i+m+hk}, \quad (i=1, \ldots, n;\ y_{j+n} \equiv y_j), \tag{5}$$

where m and h are integers. It is assumed that the system has a solution on $[a, b]$.

Theorem 2. With regard to (5), for each integer k there exists exactly one integer j_k such that $0 \leqq m+hk-nj_k \leqq n-1$. Set $s(k) = m+hk-nj_k$. Denote by $q>0$ the greatest common integral factor of h and n, and let $p = n/q$. Then $s(k)$ has the following properties:

i. $s(k+p) = s(k)$ for every k.
ii. If for some k and some positive integer $\bar{p}$, $s(k+\bar{p}) = s(k)$, then $\bar{p} \geqq p$.

iii. The set $\{s(k)\}$ of all of the values of s contains exactly p distinct elements.

Note that $s(k)$ as defined is a function whose domain of definition is the set of integers and whose range is a subset of $0, \ldots, n-1$. Also note that property ii implies that p is the least positive integer for which property i holds.

Proof of i. Because p and h/q are integers,

$$\begin{aligned} s(k) &= m+hk-nj_k \\ &= m+h(k+p)-n(j_k+h/q) \\ &= s(k+p), \end{aligned}$$

implying $s(k)$ is periodic with period p.

Proof of ii. Consider

$$s(k+\bar{p}) = m+h(k+\bar{p})-nj_{k+\bar{p}}.$$

It follows that

$$j_{k+\bar{p}} = j_k + \frac{h\bar{p}}{n}$$

and therefore that $h\bar{p}/n$ is an integer. Let $t=h/q$. Then $t\bar{p}/p$ is an integer and hence, since t and p are relatively prime, $\bar{p}/p$ is a positive integer. But this in turn implies $\bar{p} \geqq p$.

Proof of iii. From property i, $\{s(k)\}$ can have at most p distinct elements. Suppose $s(k_1)=s(k_2)$, $k_2>k_1$. Then

$$s(k_2) = s(k_1+[k_2-k_1]) = s(k_1).$$

Property ii implies $k_2-k_1 \geqq p$ so that $\{s(k)\}$ must have at least p distinct elements.

Theorem 3. Let $s(k)$ be the function defined above. Then the system

$$(6) \quad y_i' = \sum_{k=1}^{n} A_k\, y_{i+s(k)}, \qquad (i=1, \ldots, n;\ y_{j+n} \equiv y_j)$$

is identical to (5).

Proof. $y_{i+m+hk} \equiv y_{i+s(k)+nj_k} \equiv y_{i+s(k)}$.

Let $s(k)$ and p be as defined in Theorem 2. Further, let $s_1, \ldots, s_p$ denote the distinct values of $s(k)$ and suppose $s_1 < \ldots < s_p$. Each of the integers s_j satisfies $0 \leqq s_j \leqq n-1$. Functions $B_l(x)$, $(l=0, \ldots, n-1)$, are now defined in terms of s_j and the coefficients A_k of (5). This is done in the following manner:

i. If $l=s_j$, set $B_l = \sum_k A_k$,

k such that $s(k)=s_j$.

ii. If $l \neq s_j$, set $B_l=0$.

Theorem 4. The system

$$(7) \quad y_i' = \sum_{l=0}^{n-1} B_l\, y_{i+l}, \qquad (i=1, \ldots, n;\ y_{j+n} \equiv y_j)$$

is equivalent to (6). That is, every solution of (7) is also a solution of (6) and conversely.

Proof. The theorem is a consequence of Theorem 2 and the definition of the functions B_l.

Theorem 5. System (7), and hence (5), is equivalent to the matrix differential equation

$$(8) \qquad Y' = BY,$$

where

$$Y = \begin{bmatrix} y_1 \\ \cdot \\ \cdot \\ \cdot \\ y_n \end{bmatrix}, \qquad Y' = \begin{bmatrix} y_1' \\ \cdot \\ \cdot \\ \cdot \\ y_n' \end{bmatrix},$$

and B is the circulant (b_{ij}) whose elements are the coefficients of (7) and are described by

$$b_{ij} = \begin{cases} B_{j-i}, & j \geqq i \\ B_{j-i+n}, & j < i \end{cases}$$

Proof. It suffices to show that the i-th equation of (7) is identical to the i-th component of the column vector $Y'=BY$. In view of $y_{j+n} \equiv y_j$, one has

$$\begin{aligned} y_i' &= \sum_{l=0}^{n-1} B_l\, y_{i+l} \\ &= B_0\, y_i + B_1\, y_{i+1} + \ldots + B_{n-i}\, y_n \\ &\quad + B_{n-i+1}\, y_1 + \ldots + B_{n-1}\, y_{i-1} \\ &= \sum_{k=i}^{n} B_{k-i}\, y_k + \sum_{k=1}^{i-1} B_{k-i+n}\, y_k \\ &= \left(\sum_{k=i}^{n} + \sum_{k=1}^{i-1}\right) b_{ik}\, y_k = \sum_{k=1}^{n} b_{ik}\, y_k, \end{aligned}$$

which is the i-th component of $Y'=BY$.

It is now possible to give the solutions of (5). Let

$$(9) \qquad Y = TU,$$

where U is the column vector with components $u_1, \ldots, u_n$ and T is the matrix of Theorem 1. Since T is a matrix of constants,

$$Y' = TU'. \tag{10}$$

Substituting (9) and (10) into (8) and applying Theorem 1 yields

$$TU' = BTU = TDU,$$

where D is the diagonal matrix defined by (4) with $a_k = B_k$. It follows that

$$U' = DU \tag{11}$$

and hence that

$$u_i' = d_{ii}\, u_i = \Big(\sum_{k=0}^{n-1} B_k\, r_i^k\Big)\, u_i.$$

If now the functions $A_1(x), \ldots, A_n(x)$ are integrable on $[a, b]$, then so are $B_0(x), \ldots, B_{n-1}(x)$. In which case, if x belongs to $[a, b]$,

$$u_i = c_i \exp \int_a^x \sum_{k=0}^{n-1} B_k(t)\, r_i^k\, dt$$

so that

$$\begin{aligned} y_i &= \sum_{j=1}^{n} r_j^i\, u_j \\ &= \sum_{j=1}^{n} c_j\, r_j^i \exp \int_a^x \sum_{k=0}^{n-1} r_j^k\, B_k\, dt, \end{aligned} \tag{12}$$

which establishes the following theorem:

Theorem 6. If $A_1(x), \ldots, A_n(x)$ are integrable on $[a, b]$, then the solutions of (5) on $[a, b]$ are given by (12) and the solutions of (8) by $Y = (y_1, \ldots, y_n)$.

Extensions

An essential ingredient in obtaining the solutions of (5) was the function $s(k)$ of Theorem 2. The transition from (6) to (7), and then to the desired solutions, was made possible by the simple fact that $s(k)$ in (6) was a function of $k = 1, \ldots, n$ into $s = 0, \ldots, n-1$. This suggests the following generalization of (6), hence of (5). Note that n and m need not be equal.

Theorem 7. Let $\alpha(k)$ be any function having as its domain of definition the integers $1, \ldots, m$ and as its range of values a subset of $0, \ldots, n-1$. Denote by $\alpha_1 < \ldots < \alpha_p$ the distinct values of $\alpha(k)$. Let $A_1(x), \ldots, A_m(x)$ be integrable functions on $[a, b]$. Further, let $B_{\alpha_j} = \sum_k A_k$, k such that $\alpha(k) = \alpha_j$, and $B_h = 0$, $h \neq \alpha_j$. Then the solutions of the system

$$y_i' = \sum_{k=1}^{m} A_k\, y_{i+\alpha(k)}, \qquad (i = 1, \ldots, n;\ y_{j+n} \equiv y_j) \tag{13}$$

are given by (12).

Proof. As in the argument leading to Theorem 6, show that (13) is equivalent to $Y' = BY$, where B is the circulant with elements B_j, and apply Theorem 1 to obtain (11), then (12).

In order to clarify the proof of Theorem 7 and, at the same time, illustrate the use of (12), the following example is treated in detail. Note that the example is not included among those systems described by (1).

Example. With regard to (13), take $n = 3$, $m = 4$, and $\alpha(k) = k^2 \pmod 3$. Then, because $\alpha(1) = 1$, $\alpha(2) = 1$, $\alpha(3) = 0$, $\alpha(4) = 1$, and $y_{j+3} \equiv y_j$, the equations of (13) assume the form

$$\begin{aligned} y_1' &= A_1y_2 + A_2y_2 + A_3y_1 + A_4y_2 \\ y_2' &= A_1y_3 + A_2y_3 + A_3y_2 + A_4y_3 \\ y_3' &= A_1y_1 + A_2y_1 + A_3y_3 + A_4y_1 \end{aligned}$$

which in turn may be rewritten

$$\begin{aligned} y_1' &= B_0y_1 + B_1y_2 + B_2y_3 \\ y_2' &= B_2y_1 + B_0y_2 + B_1y_3 \\ y_3' &= B_1y_1 + B_2y_2 + B_0y_3 \end{aligned} \tag{14}$$

where

$$\begin{aligned} B_0 &= A_3 \\ B_1 &= A_1 + A_2 + A_4 \\ B_2 &= 0. \end{aligned}$$

This system is equivalent to $Y' = BY$, where B is the circulant

$$\begin{bmatrix} B_0 & B_1 & B_2 \\ B_2 & B_0 & B_1 \\ B_1 & B_2 & B_0 \end{bmatrix} = \begin{bmatrix} A_3 & A_1+A_2+A_4 & 0 \\ 0 & A_3 & A_1+A_2+A_4 \\ A_1+A_2+A_4 & 0 & A_3 \end{bmatrix}.$$

Direct substitution of

$$y_j=\sum_{k=1}^{3} r_k^j u_k,\ (j=1, 2, 3),$$

in (14) with the cube roots r_k of unity given by

$$r_k=\cos\frac{2k\pi}{3}+i\sin\frac{2k\pi}{3},\ (k=1, 2, 3),$$

yields, upon solving for u_1', u_2', u_3',

$$u_j'=(B_o+B_1r_j+B_2r_j^2)\,u_j,$$

whereupon, if $A_1, \ldots, A_4$ are integrable,

$$u_j=c_j\exp\int_a^x (B_o+B_1r_j+B_2r_j^2)\,dt$$

and hence

$$(15)\quad y_j=\sum_{k=1}^{3} c_k r_k^j \cdot \exp\int_a^x (B_o+B_1r_k+B_2r_k^2)\,dt.$$

Having determined B_o, B_1, B_2 and the cube roots of unity, this same result could have been obtained immediately from (12). That the solution so obtained does indeed satisfy the given system is easily verified by direct substitution in (14).

Theorem 8. Let $\alpha_l(k)$, $(l=1, 2, \ldots)$, be functions of the positive integers into 0, $\ldots, n-1$. Denote by $\bar{\alpha}_1<\ldots<\bar{\alpha}_p$ the distinct values of $\alpha_l(k)$, $(k, l=1, 2, \ldots)$. Let $A_{kl}(x)$, $(k, l=1, 2, \ldots)$, be functions on $[a, b]$ with the property that $\sum_{k,l} A_{kl}(x)$ converges absolutely and uniformly to an integrable function on $[a, b]$. Further, let $B_{\alpha_j}=\sum_{k,l} A_{kl}$, k and l such that $\alpha_l(k)=\bar{\alpha}_j$, and $B_h=0$, $h\neq\bar{\alpha}_j$. The solutions of the system

$$y_i'=\sum_{k,l} A_{kl}\,y_{i+\alpha_l(k)},\quad (i=1, \ldots, n;\ y_{j+n}\equiv y_j),$$

are given by (12).

Proof. Same as Theorem 7, keeping in mind the implications of the convergence of $\sum_{k,l} A_{kl}$.

Example. Take $n=3$ and let

$$\alpha_1(k)=1+(-1)^k,\quad \alpha_2(k)=1,$$

and $\alpha_l(k)$ be arbitrary for $l>2$. Define $A_{kl}=0$ for $l>2$ and

$$A_{k1}=\frac{x^{k-1}}{(k-1)!},\quad A_{k2}=2^{-k}.$$

With these agreements the system of Theorem 8 becomes

$$y_i'=\sum_{k=1}^{\infty} A_{k1}\,y_{i+\alpha_1(k)}+\sum_{k=1}^{\infty} A_{k2}\,y_{i+\alpha_2(k)},$$
$$(i=1, 2, 3;\ y_{j+3}\equiv y_j),$$

the first equation of which is

$$y_1'=y_1+xy_3+\frac{x^2}{2!}y_1+\frac{x^3}{3!}y_3+\ldots$$
$$+\tfrac{1}{2}y_2+\tfrac{1}{4}y_2+\tfrac{1}{8}y_2+\ldots$$
$$=y_1\cosh x+y_2+y_3\sinh x.$$

From this it is seen that the second and third equations are

$$y_2'=y_1\sinh x+y_2\cosh x+y_3$$
$$y_3'=y_1+y_2\sinh x+y_3\cosh x$$

so that $B_o=\cosh x$, $B_1=1$, $B_2=\sinh x$. Thus, when these values together with $n=3$ and the cube roots of unity are substituted in (12), the solution assumes the form of (15).

Although Theorems 7 and 8 were regarded as separate theorems, for reasons of presentation, Theorem 7 is in fact a corollary of Theorem 8. This is easily seen if one sets $A_{kl}=0$, $l\neq 1$, $A_{k1}=A_k$, and $\alpha_1(k)=\alpha(k)$.

It was assumed in Theorems 6, 7, and 8 that A_k and A_{kl} were real functions of the real variable x. Inasmuch as the elements of the circulant A of Theorem 1 may be complex valued functions of a real or complex variable, the results of these theorems are easily extendible to cyclicly related equations whose coefficients are complex functions.

LITERATURE CITED

1. BROWN, E. T. Introduction to the Theory of Determinants and Matrices. University of North Carolina Press: Chapel Hill, N. C., 1958.
2. WHYBURN, W. M. A Set of Cyclicly Related Functional Equations. Bull. Amer. Math. Soc. **36**: 863–868. 1930.

INTERRELATION OF FUNCTIONAL MORPHOLOGY, BEHAVIOR, AND AUTECOLOGY IN EARLY STAGES OF THE BIVALVE *MERCENARIA MERCENARIA*

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Introduction

The hard clam or quahog, *Mercenaria mercenaria* (Linne 1758) (formerly *Venus mercenaria* Linne), Family Veneridae (Frissel, 1936; Turner, 1953; H. Wells, 1957; Aldrich, personal communication) principally inhabits the shallow, more saline coastal waters along the eastern and gulf coasts of North America from Nova Scotia to Yucatan and is harvested in large quantities as a wild crop. Although adult stages of the species have received considerable study (see review, Carriker, 1959), the larval stages have been given much less attention, and efforts to propagate the bivalve on a commercial scale are only now beginning to be successful. Early stages in the life history of the venerid are most vulnerable to depredations of enemies, yet their study in the laboratory and in the field has been possible only within the last decade or so during which laboratory culture of the larvae has been accomplished (Loosanoff, 1954).

In memory of

Thurlow Christian Nelson

This paper reports results of studies on, and interrelation of, functional morphology, behavior, and autecology of swimming and early bottom-dwelling stages of *Mercenaria mercenaria* in the laboratory and in the field. These studies were made by the author and associates during the period from 1947 to 1956 in New Jersey, New York, and North Carolina. Before 1950 the only report of consequence on the biology of the early stages of *M. mercenaria* in the literature was by Belding (1912), who reported briefly on the general embryology of the larvae through the straight-hinged stage and on the general anatomy and habits of the byssal stage. He was able to raise the larvae only to the straight-hinged stage and obtained newly set byssal stages in open boxes in the field. In the last decade Loosanoff and associates have made substantial contributions to knowledge of the culture and physioecology of the larvae in the laboratory (Loosanoff, 1953, 1954, 1958, 1959b; Loosanoff et al., 1950, 1951a, 1951b, 1953a, 1953b; Davis, 1958; Davis et al., 1956, 1958; Chanley, 1958; Lindsay and Woelke, 1960); and Carriker (in both laboratory and field, 1950b, 1952, 1956) and Turner and George (in the laboratory, 1955) have reported results of preliminary studies on the behavior of pelagic and early post-set stages.

To make accurate identification of veligers of *M. mercenaria* in native waters possible and to supply a large quantity of veliger and post-set stages for laboratory investigations, we reared young individuals under field laboratory conditions. We were aided substantially by larval-culture techniques which Loosanoff and associates were developing at the time and generously shared with us.

For the most part we conducted our investigations during the summer months. In New Jersey (1947–1951) laboratory facilities and living accommodations were provided by the Oyster Investigation Floating Laboratory, the "Cynthia," a houseboat which we moored each summer in a tidal creek adjacent to the body of water in which field studies were made (Carriker, 1961). Tide level, salinity, water and air temperatures, barometric pressure, rainfall, wind, and cloudiness of the sky were recorded routinely from the "Cynthia" to provide information used in connection with study of spawning, culture, behavior, and larval distribution and movements of the hard clam. A twenty-six-foot flat-bottomed cabin boat (a New Jersey "garvey") with inboard engine and equipment for pumping plankton samples, dredging, and washing sediment samples facilitated field work.

In the summer of 1947 we moored the "Cynthia" in Landing Creek and conducted field studies in Great Bay (Fig. 1). This estuary is six statute miles long and four miles wide

[a] Now at the U. S. Bureau of Commercial Fisheries Biological Laboratory, Oxford, Md.

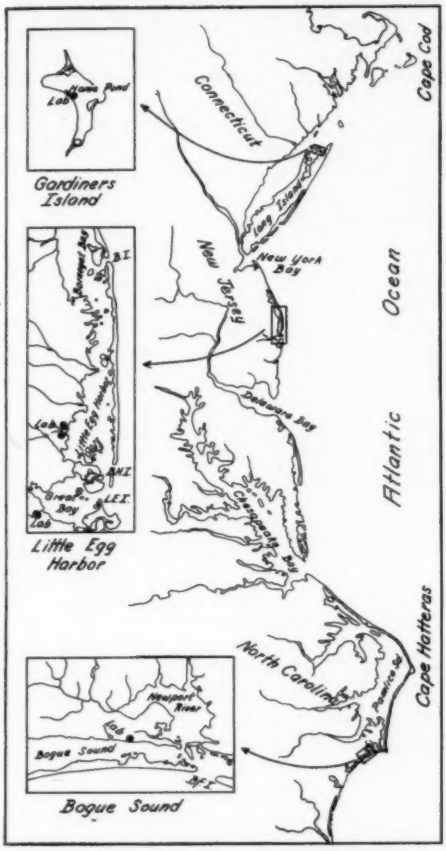

FIG. 1. Chart of east coast of the United States from Cape Cod to Cape Fear, showing location of study areas and magnified insets of these areas: Little Egg Harbor, New Jersey, 1947–1951; Gardiners Island, New York, 1953–1955; Bogue Sound, North Carolina, 1956. BI, Barnegat Inlet; BfI, Beaufort Inlet; BHI, Beach Haven Inlet; LEI, Little Egg Inlet.

and has an average depth at mean low water of six feet. It opens to the Atlantic Ocean through numerous deep channels by way of Little Egg Inlet and receives large volumes of fresh water from the Mullica River. Maximum current velocities in the middle of the estuary in 1947 approximated 0.8 ft./sec. Spring range of the tide was four feet, summer salinities in the middle of the bay ranged from 18 to 31 °/oo, and water temperatures from 21° to 30°C. Sediments varied from clean fine sand near the inlet to highly organic mud in the quieter upper reaches of the estuary. Sparse populations of *M. mercenaria* were scattered over the lower reaches, providing clams for limited commercial tonging. Carriker (1951b) describes the hydrography and fauna of this bay more fully.

We moored the "Cynthia" in Ezras Creek in the summer of 1948 and in Thompson Creek in the summers of 1949–1951. Both are small tidal creeks draining across a salt marsh one to three miles in width eastward into Little Egg Harbor, where we conducted field studies (Fig. 1) (Carriker, 1961). A small cabin beside Thompson Creek became an annex for culture of larvae of *M. mercenaria*. No running sea water was available in our laboratory. In contrast with Great Bay, Little Egg Harbor receives only a small flow of fresh water from small streams, and, in spite of continued harvesting by local clammers, at the time of our studies was inhabited by dense populations of *M. mercenaria*. The detailed hydrography of Little Egg Harbor is treated in a subsequent section on the ecological study of larvae of *M. mercenaria* in the field.

No field studies were conducted in 1952. During the summers of 1953–1955 we made our investigations on Gardiners Island, New York (Fig. 1). A small cabin equipped with running sea water on the eastern shore of Home Pond provided our laboratory facilities. The sea water system consisted of a hard rubber pump, lead and hard rubber pipes, and a wooden reservoir. A heating unit (sea water passed through lead coils in tap water heated by gas flame) in the system heated sea water for artificial spawning of *Mercenaria*, and we had a small rowboat with an outboard motor for making field studies in Home Pond and the other three large ponds on the island. Home Pond is a shallow open-inlet 21-acre arm of Gardiners Bay with a maximum amplitude of 2.4 feet. Fresh water entering the pond comes in as rainfall and seepage from surrounding forested slopes. During the three summers salinities ranged from 22 to 33 °/oo in the pond, while those in Gardiners Bay seldom fell below 28 °/oo. The bottom of the outer half of the pond consisted of clean sand; the inner half, fine organic mud. There were dense populations of *M. mercenaria* in the pond, especially in the inlet and vicinity; these were not harvested commercially. See Carriker (1959) for a description of the hydrography of Home Pond and vicinity.

In the summer of 1956 research shifted to the University of North Carolina Institute of Fisheries Research in Morehead City (Fig. 1) on Bogue Sound. Here a study of the culture and behavior of young *Mercenaria* in the labora-

tory received prime emphasis. Constantly running sea water was pumped (hard rubber pump) to and distributed in the laboratory in non-toxic plastic (polyethylene) pipe (Carriker, 1957). Bogue Sound is a long, narrow, shallow coastal-plain lagoon separated by a narrow barrier island from the Atlantic Ocean to which it communicates by an inlet at each end. Most, if not all, of the sea water off the laboratory entered the sound through Beaufort Inlet. The sound receives relatively little fresh water; salinities, which fluctuated slightly from week to week, rose from 31 to 36 °/oo during the course of the summer. Temperature of the sea water entering the laboratory varied between 23° and 31°C., but remained above 27°C. most of the season. *M. mercenaria* occurred in moderate abundance in Bogue Sound (Chestnut, 1952) and supported a small commercial fishery.

I LABORATORY CULTURE OF YOUNG *MERCENARIA*

When we began our studies in 1947 there was very little information available in the literature on the biology of the early stages of *M. mercenaria*, and identification of the larva was in doubt. The papers of Stafford (1912), Belding (1912), Nelson (1921), and Wells (1927) provided illustrations and in some cases descriptions, but these were not in agreement. Stafford and Nelson attempted identification by tracing development of successively older stages in the plankton. Belding achieved fertilization of ova and grew larvae to the straight-hinged stage in the laboratory. Wells, the first to carry larvae to the late stage in the laboratory, published good photomicrographs of them but omitted descriptions; unfortunately identification of larvae from illustrations alone is difficult and unreliable. Comparable stages of the larvae illustrated by Wells and by Belding resemble each other, but not those of Stafford and Nelson.

During the summer of 1947 we made negligible progress in identifying the larvae of *M. mercenaria*, since our laboratory-reared larvae developed only to the trochophore stage. In 1948 we reared them to a length of 142 μ. Concurrently Nelson, Haskin, and Gibbs, at the Cape May laboratory on Delaware Bay in New Jersey, grew them to a setting size of 208 μ; and during the winter of 1948–1949 Loosanoff (personal communication; Loosanoff and Davis, 1950) for the first time cultured them to the setting stage in large numbers. Culture of larvae of *M. mercenaria* in the laboratory from gametes of known parents thus made possible positive identification of these larvae in plankton of native waters (Carriker, 1950b). Our larvae resembled those described by Belding (1912) and by Wells (1927) and those reared by Loosanoff. At the same time Sullivan (1948) published a description and illustrations of a larva which she misidentified as *Mercenaria mercenaria* by tracing development of larval stages in the plankton. This larva resembles the one described by Nelson (1921) as *M. mercenaria* and is remarkably similar to an unidentified bivalve larva which we found in moderate abundance in Great Bay, Little Egg Harbor, and Bogue Sound, and which during the summer of 1947 we too mistook for the larva of *M. mercenaria*.

Profiting from the larval culture technics of Loonsanoff and associates (personal communications; Loosanoff and Davis, 1950, 1951a; Loosanoff, 1953, 1954; Loosanoff et al., 1953a, 1953b), we reared larvae through post-setting stages during the summers of 1951 in Little Egg Harbor, 1953–1955 on Gardiners Island, and 1956 in Morehead City. (This last represents the first time that larvae of *M. mercenaria* have been cultured successfully in the laboratory in the southeastern United States [Carriker, 1957].) We developed routine methods for collection, preparation, and artificial stimulation of adults for spawning; for artificial fertilization of ova; and for cleaning and feeding and observing planktonic and post-set stages of *M. mercenaria*. In the application of these procedures we made several observations on the requirements and culture of the larvae, some of which are reported here. Part of these confirm similar observations reported by Loosanoff and associates and by Turner and George (1955), and others are recorded for the first time.

Spawning

Of 2,890 individuals of *M. mercenaria* placed in spawning trays during the course of six summers, 12 per cent spawned, a result close to the 17 per cent figure obtained by Loosanoff (1937) in similar laboratory spawnings.

Clams were dug by hand, raked, or tonged, and a sample opened for macro- and microscopic examination of gonads and gametes. Potential spawners were then brushed and washed thoroughly and arranged in trays of still or running

clean sea water of a salinity approximating that of sea water at high tide where the clams were collected. If temperature of the water was below 23°C., it was raised to about 28°C. by immersion of the spawning trays in larger trays of warm water or by heating the running sea water directly. Temperatures much above 28°C. caused many clams to pump more slowly, or even to close. After the clams had been thermally conditioned for an hour or so and most were pumping actively, we added a few cubic centimeters of a dense suspension of active sperm to the water in the vicinity of the incurrent siphon of each clam. (In a number of bivalves, including the hard clam, spawning is a group reaction stimulated by a hormone carried on the sperm [Nelson and Haskin, 1949].) If necessary, subsequent additions were made from time to time. Sperm were obtained by stripping males with well-developed gonads: integument was sliced from the gonads of several individuals, and with a bulb pipette enough sperm were teased and flushed from the gonads into a beaker of filtered sea water to provide a milky white suspension. We took care to avoid cutting the digestive diverticulum in order to prevent introduction of digestive enzymes. Tissue fragments were removed from the suspension by decantation. Only active motile sperm were utilized; the degree of activity was checked under a magnification of 440X, at which the slightly curved sperm heads, 6 to 8 μ in length, with their long flagella, could be seen whirling in small circles. Stripped sperm in a dense suspension were active as long as six hours at between 22° and 24°C. Loosanoff (1954) has shown that viability of gametes of *Mercenaria* is unrelated to size or age of parents, and we therefore selected parents at random from those available.

A common response of *Mercenaria* to the addition of sperm, particularly when ready to spawn, was to open the siphons more widely and pump more vigorously. Ready-to-spawn (physiologically mature) clams started spawning five minutes to one hour after addition of sperm, males invariably spawning before females (also see Loosanoff, 1937). Refractive individuals did not spawn for several hours.

Clams siphoned and responded more actively to sperm stimulation in water which had been warmed from a temperature below 25°C. or cooled from a temperature above 28°C. (also see Loosanoff and Davis, 1950; Turner and George, 1955) than in water of a constant high temperature; and there was also a more active response in running sea water than in still sea water. Because of the high temperature of the water entering the laboratory at Morehead City from Bogue Sound (27° to 31°C. much of the summer) warming the water to induce spawning was unnecessary. The spawnings that occurred were obtained in the late afternoon and evening as water and air temperatures dropped, and several hours after the addition of the sperm. Infrequently in all areas clams spawned as a result of thermal change alone.

Spawning of the first male in any tray soon started other males spawning, and later the females also. These individuals were then moved gently to separate containers of filtered sea water to complete spawning.

Emission of gametes continued in spite of such molestations as jarring of the container, partial change of water by dipping or siphoning, and movement of the spawner to another container. Duration of continuous emission was variable depending, among other factors, upon the fullness of the gonad and recent spawning history. Maximum observed continuous emission for females was 10 minutes, and for males 35 minutes.

Gametes are voided through the excurrent siphon. In males, sperm are emitted in a white thread-like stream which frays out of macroscopic vision some 1.5 to 2 inches from the siphon tip; in females ova are generally released in a steady loose granular white spray which fans broadly just outside the siphon tip, and since ova measure approximately 125 μ in diameter (including a clear gelatinous envelope; also see photograph, Loosanoff and Davis, 1950) they remain visible macroscopically. Occasionally females voided ova in the form of pellets which accumulated beneath the siphon.

Conditioning and spawning temperature of the water ranged between 25° and 28°C. The maximum temperature of induced spawning was 29°C. and minimum temperature, 23°C. This minimum, which was associated with a spawning on Gardiners Island, was the prevailing temperature for some hours before the addition of sperm to the spawning tray. H. J. Turner and J. Hanks (personal communication) in early June in Massachusetts spilled a small quantity of sperm and ova into a water table where spawning stocks of *Mercenaria* were being maintained in water at 19°C. A wave of mass spawning started at the upper end of the water table and progressed through the population for fifteen feet. This is the lowest temperature recorded for spawning in the laboratory; note that Loosanoff (1951b) has shown that newly fertilized ova fail to develop normally at temperatures of 15°C. and below.

Table I

Frequency of spawning of *Mercenaria mercenaria* in the laboratory in New Jersey, New York, and North Carolina during the summer months, 1948–1956.

Periods spawning attempted	New Jersey				New York						North Carolina			
	1948		1951		1953		1954		1955		1956		1956 Va.-refr.	
	No. clams used	Clams spawned, %	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
June 1–10	—	—	—	—	—	—	—	—	—	—	144	1	72	14
11–20	—	—	—	—	—	—	13	23	31	58	48	38	13	85
21–30	—	—	82	5	—	—	23	86	—	—	80	0	10	10
July 1–10	49	0	89	1	86	20	82	78	147	5	—	—	9	78
11–20	72	24	278	15	68	6	23	4	—	—	40	0	23	9
21–31	78	26	89	1	40	8	60	53	221	0	—	—	—	—
Aug. 1–10	—	—	284	1	22	27	—	—	—	—	40	0	9	33
11–20	48	0	102	0	49	16	18	78	265	1	—	—	—	—
21–31	—	—	74	4	10	90	—	—	—	—	—	—	—	—

Frequency of spawning in the laboratory varied somewhat in the three geographic areas (Table I). Most abundant and consistent spawnings were obtained in 1954, and minimal success was achieved with North Carolina clams in 1956. In New Jersey the peak of spawning came in July; in New York good spawnings were obtained over much of the summer during the first two years, but only in June of the last summer. In North Carolina *Mercenaria* had spawned partially to completely before June 1–8, when a survey of the condition of gonads in clams from different areas was made. Gonads of clams from warmer waters of Core Sound, Black Sound, and Bogue Sound were more nearly depleted than those of clams from bottoms near inlets. Between June 1 and August 9, 352 clams from these sounds were placed repeatedly in spawning trays; one spawned on June 5 and twenty-one on June 12, and thereafter no further gametes were obtained.

RETARDATION OF SPAWNING. Because frequently the bulk of spawning occurred early in the season, it became increasingly difficult to induce spawning as the summer season advanced. Retardation of spawning by retention of clams at low temperatures (Loosanoff and Davis, 1951a) helped to circumvent this difficulty and made possible the rearing of larvae in Morehead City, where zygotes from local *Mercenaria* never developed. We had observed, as did Loosanoff (1937), that clams in shallow water begin to breed earlier than those in the deeper, colder parts of the sounds, especially near inlets. Thus on June 15, 1953, we harvested unspawned *Mercenaria* from deep water off Greenport, Long Island, and kept them in a cold room at approximately 15°C. in shallow enamel trays of sea water which was changed weekly. As late as eight weeks after harvesting, they spawned profusely when stimulated by sperm and increased temperatures. Their larvae developed normally to the setting stage. After nine weeks in the cold room, most individuals spawned fully, but larvae were abnormal and did not metamorphose.

In Morehead City we obtained a supply of unspawned *Mercenaria* from Cheasapeake Bay, Virginia, on June 1. These we housed in a refrigerator at between 5° and 10°C. in trays of still sea water, except for a daily period of an hour when they were immersed in warm running sea water in the laboratory to feed. Larvae from clams maintained under these conditions over the period of a month developed normally to the setting stage, but veligers from animals kept thus longer than a month displayed anomalies.

Loosanoff and Davis (1951a) delayed spawning of clams and made available viable gametes through the summer and fall by transferring individuals with physiologically mature gonads in May from Long Island Sound to Boothbay Harbor, Maine, where water temperatures

in the summer were between 5° and 8°C. lower than in Long Island Sound.

Fertilization

Fertilization of ova was accomplished in large shallow trays by addition of a few cubic centimeters of sperm suspension for each liter of dense ova suspension. Frequently the number of sperm used to induce spawning was sufficient to fertilize all ova obtained. Use of excess sperm resulted in abnormal development of embryos. In all three geographic areas fully spawning, vigorous females that eliminated ova quickly and abundantly produced the most vigorous, fastest-growing, most viable larvae. Embryos developing from ova released in pellets, or from ova released some time after stimulation and in small quantities, often did not metamorphose. Satisfactory development of embryos resulted from fertilization of naturally released ova by either active stripped or naturally released sperm. Fertilized stripped ova never developed (also Loosanoff, 1937).

Culture Methods

As soon as the first polar body was evident, embryos were washed gently with clean cotton-filtered sea water through a three-inch stainless steel screen No. 100 (pore size 165–210 μ) onto a second screen No. 325 (pore size 45–65 μ) to remove excess sperm, mucus, fecal strings which had accumulated in the spawning tray, and tissue fragments and blood from the stripped gonads and were stored in shallow trays of filtered sea water. In less than 12 hours embryos developed to active ciliated gastrulas, and in 16 to 24 hours into early straight-hinged veligers. Twenty-four-hour-old veligers were then transferred to glass or earthenware crocks, ranging in volume from 1 to twenty liters, in concentrations averaging from 5 to 10 larvae per cubic centimeter of sea water. Densities of 50 larvae per cubic centimeter were raised to metamorphosis in 11 days in 1951, but in general larvae in concentrations much above 10 per cubic centimeter grew less rapidly and were subject to higher mortality. Loosanoff et al. reported (1953a) similar observations. Richards (1933) reported that stainless steel screens are nontoxic when used in the culture of *Mytilus edulis* ranging in length from 5–8 mm. over a period of 41 days in still sea water changed every third day; their use in the culture of clam larvae by Loosanoff and associates and by us confirms Richards' observations.

In early culturing efforts we placed hard-clam larvae in glass tunnels covered at each end with silk bolting cloth and suspended in the water over the side of the "Cynthia" in Thompson Creek. However, insufficient exchange of sea water and lack of food prevented development much beyond the straight-hinged stage. This failure led during the summer of 1951 to the use of the Browne plunger jar (Fig. 2 [Browne, 1897; Lebour, 1933; Bruce et al., 1940; Lebour, personal communication]), in which we reared several broods past the setting stage. In the meantime Loosanoff et al. (1951b) found that aeration of cultures in unnecessary. Consequently, after 1951 we employed only still-water cultures without supplementary aeration of any kind.

FIG. 2. Plunger jars and oscillating frame employed in culture of *M. mercenaria* during summer of 1951 in Mott's cabin, Thompson Creek, Little Egg Harbor, New Jersey.

Approximately every two days the sea water in the larval culture jars was replaced completely with cotton-filtered sea water collected at high tide. When sea water from a laboratory running-sea-water system was used, presence or absence of deleterious metals and other toxic substances in the system was checked by rearing larvae concurrently in controls consisting of sea water hauled manually in enamel or glass containers. During water changes larvae were siphoned onto a three-inch stainless steel screen through rubber tubing held securely to the sides of the screen by means of a large one-hole rubber stopper. The culture jar was washed and the residue rinsed onto a second screen to retain larvae remaining in the residue. As larvae grew in size we used successively coarser screens No. 250 (maximum pore size [that is, diagonal dimension of square opening], 110 μ) and No. 150 (pore size, 150 μ) to retain them. Fresh food microorganisms were added after each change of sea

water, and daily if the larvae cleared the water of planktonic algae during the previous day.

After larvae metamorphosed and began creeping on the bottom of the culture jar, we removed only about four-fifths of the water at each change, since individuals at this stage adhere tenaciously to substrata and are readily transported on screens to contaminate other cultures when the same screen is employed in cleaning several jars. To eliminate the possibility of such transference we dipped screens in fresh water after each change. This causes clams to withdraw the foot between the valves and permits flushing them from screens. The practice of partial removal of water left detritus on the bottom of culture jars which (see page 189) provided a medium for burrowing by older stages.

Larvae survived air temperature fluctuations ranging between 13°C. and 30°C. in our laboratories.

*p*H of the sea water in larval cultures usually fluctuated between 7.8 and 8.0.

SUPPLEMENTAL FEEDINGS. Food micro-organisms were first added to each new larval culture soon after the straight-hinged stage was reached. We did not have an opportunity to determine whether growth of preshelled stages could be accelerated by addition of glucose and/or amino acids which might become available to them through absorption from sea water, but this should be investigated. In Little Egg Harbor and on Gardiners Island approximately 35 cubic centimeters of a dense suspension of algal cells were added to each liter of sea water containing 50 larvae per cubic centimeter; correspondingly lesser quantities were contributed to sparser larval cultures. In 1955 and 1956 algal cells were added in quantities recommended by Loosanoff et al. (1953b) and a haemocytometer was used to determine algal concentrations.

Attempts during the summers of 1947 and 1948 to raise larvae without supplemental feeding—that is, relying on a daily or twice daily change of sea water to provide necessary nutrients—were unsuccessful; at best larvae could be carried to a maximum length of only 170 μ before they died in 12 to 18 days.

During the last summer in Little Egg Harbor and the three summers on Gardiners Island, supplemental food of mixed micro-organisms dominated by *Chlorella* sp. (young cells averaged 3 μ in diameter), cultivated by a method modified from that recommended by Loosanoff and Davis (1950), was provided routinely to many of our larval cultures.[1] On the whole, however, in contrast with the success of Loonsanoff and Davis (1950), we were able to raise hard clam larvae to setting on mixed culture of *Chlorella* in only occasional larval cultures; and in these setting did not start until the fourteenth day after fertilization, and growth was accompanied by excessive mortality.

In Little Egg Harbor in 1951 optimum results in growing hard-clam larvae were obtained by feeding them a mixed culture inoculated with *Chlorella* cells and fertilized heavily (10 g. per liter) with 6–3–6 organic fertilizer alkalized with marble chips. These food cultures passed through a succession of several colors and odors over a period of from three to four weeks before they were fed to the larvae. Both whole culture and filtrate (from Whatman filter paper No. 42, which probably permitted a sizeable fraction of smallest micro-organisms to pass) raised larvae to post-setting stages in large numbers. Fastest-growing larvae reached setting size in ten days, with very little mortality. On Gardiners Island, however, *Chlorella* cultures fertilized with 2, 3, and 10 grams of fertilizer per liter, as well as filtrates of these, gave results inferior to or similar to those obtained with the standard mixed culture of *Chlorella* fertilized with 1 gram per liter.

Maximum survival and setting of larvae on Gardiners Island was achieved by adding pab-

[1] Food organisms were cultivated in sixteen-liter (five-gallon) wide-mouth glass jars in cotton-filtered sea water collected at high tide, and were fertilized with 6–3–6 or 5–3–5 organic fertilizer in a concentration of 1 g. per liter. Jars were agitated at least three times daily, and cells clinging to inner surfaces of jars were scraped free to permit maximum penetration of daylight, up to 400 foot-candles near screened laboratory windows. When cells in the first jar produced a dense green color (2,000,000 to 7,000,000 cells per cc.), a four-liter inoculum from culture was sufficient to produce a dense population in a new sixteen-liter culture in two or three days. Dense cultures were harvested at the rate of two liters of suspension per day; sea water was replenished as suspension was withdrawn, and fertilizer was added at the rate of 0.5 g. per liter of suspension removed. Best cultures were harvested almost continuously for as long as a month and provided a continuous crop of young cells. Occasionally some of the cultures turned yellow from insufficient light (during overcast days, light in the laboratory fell below 100 foot-candles on Thompson Creek and Gardiners Island, where no electricity was available), from dense growth of ciliates and naked flagellates which fed on the cells of *Chlorella*, or from our own overharvesting.

lum extract to the culture water. Fifteen cc. by volume of commercial cereal pablum flakes were soaked in 100 cc. of sea water for two hours and filtered through Whatman paper No. 41; one-tenth of a cc. of this filtrate was added over a period of four days to each liter of larval culture every second day following the change of sea water; subsequently 0.5 cc. of filtrate was added every second day. This quantity of filtrate, accelerating growth of bacteria and other micro-organisms, raised larvae in duplicate cultures to the setting stage in fourteen days. Larval cultures became slightly milky in appearance a few hours after addition of the filtrate. Even with this method considerable mortality of larvae still occurred.

Feeding mixtures of pablum filtrate and mixed culture of *Chlorella* to larvae demonstrated that they tolerated higher concentrations of the pablum filtrate (up to 2 cc. per liter) in the presence of the *Chlorella*, but otherwise produced variable results, associated probably with the nutritive value of the mixed culture of *Chlorella*. For example, in one series of larval cultures we could not rear larvae on mixed culture of *Chlorella* alone beyond a size of 156 μ, yet in combination with pablum filtrate, metamorphosis was obtained in ten days with relatively little mortality. Conversely, in a second series, pablum filtrate, when added with mixed culture of *Chlorella*, produced no difference in growth or survival rate from that rendered when it was added alone. In a few larval cultures, mixed culture of *Chlorella* retarded growth rate of the larvae. During the four summers in Little Egg Harbor and on Gardiners Island, a total of 111 larval cultures were started, and metamorphosis occurred in 19 per cent of these.

We obtained our best results in rearing larvae of *M. mercenaria* in Morehead City, where in the course of the summer we established thirty cultures. Seventy-seven per cent of these rendered abundant larvae which reached the post-setting stage. We prepared a variety of larval foods to insure at least one which would permit cultivation of large populations of metamorphosed clams. Among them were unialgal cultures (not bacteria free) of *Chlorella*, *Nannochloris*, and *Chlamydomonas* which we established in cotton-filtered sea water, in heat-treated cotton-filtered sea water (about 70°C. for 30 minutes), in heated cotton-filtered sea water treated with bone charcoal, and in autoclaved sea water. Algal cultures were placed in three-gallon glass jars under aeration in a porch which faced north and received from 400 to 800 foot-candles of daylight on sunny days, being shaded with screens on the east and west. One *Chlorella* culture was fertilized with 5–3–5 organic fertilizer (Loosanoff and Engle, 1942), the others with Rice's (1953) modification of Miquel's solutions. In spite of high summer temperatures these species of algae multiplied rapidly, and all cultures except those in autoclaved sea water soon contained dense mixed populations of *Nitzschia*, naviculoids, and other contaminating micro-organisms which entered in sea water. Sea water in Bogue Sound, particularly that in the eastern end, which was moderately polluted and provided the source of our laboratory running sea water, contained so high a content of nutrients that addition of one gram per liter of 5–3–5 organic fertilizer caused the usual bloom of micro-organisms attributed in our New Jersey studies to overfertilization.

Of the species of algae used, cells of *Nannochloris* fertilized with Miquel's solutions grew most rapidly, reaching concentrations of 20,000,000 cells per cubic centimeter in short periods.

We also used Fleischmann's "active dry" yeast in sea water, cooked soluble starch (1 mg. per liter), and pablum filtrate as food.

Hard-clam larvae grew most rapidly when fed mixed culture of either *Nannochloris* or *Chlorella*. On a diet of one or the other alone, they started to set in eight days. Least mortality occurred on a diet of *Nannochloris*. Post-set clams also grew well on these two mixed foods. However, unialgal cultures of *Nannochloris* cultivated in autoclaved sea water under sterile conditions produced an inferior food on which the larvae grew poorly or not at all. Good growth was also achieved by larvae fed on pablum filtrate, starch, yeast, and *Chlamydomonas* cells; these started setting in from eight to thirteen days, but considerable mortality took place. The addition of pablum filtrate to mixed culture of *Nannochloris* increased total shell length of the fastest-growing larvae in laboratory sea water by about 100 μ in twenty days, though this growth rate did not approach that by larvae reared in water hauled from the sound and fed mixed culture of *Nannochloris* alone.[2] (Faster-growing larvae

[2] The effect of different kinds of sea water in our culture of larvae was tested as follows. Duplicate cultures of hard-clam larvae fed on a mixed culture dominated by *Nannochloris* were grown in (a) sea water hauled in enamel buckets from Bogue Sound, (b) sea water taken from the laboratory running-sea-water system, (c) laboratory sea water heated to 70°C. for 30 minutes, and (d) sea water heated to 70°C. for 30 minutes

raised under the latter conditions reached lengths ranging from 0.88 to 1.10 mm. in twenty-seven days.)

Although mixed algal cultures are not always reliable, since occasionally a culture becomes dominated by an undesirable micro-organism, they afford an easily cultured nutritious larval food. Recently Davis and Guillard (1958) demonstrated that *Chlorella* constitutes a less nutritious food for larvae than *Monochrysis lutheri* and *Isochrysis galbana*. Thus it would appear, as also suggested by the low nutrient value of unialgal populations of *Nannochloris* in our cultures, that other micro-organisms than *Nannochloris* and *Chlorella* were being utilized by our clam larvae. Turner (1955) found *Nitzschia closterium* a suitable food for *M. mercenaria* larvae raised in his laboratory. This species was easily cultivated in quantity and lacked toxic properties even in high densities.

A singular lack of uniformity of results in culturing larvae of *M. mercenaria* to metamorphosis under apparently similar conditions within a single season and in different seasons and in different regions[3] led us to the hypothesis that an unidentified substance(s) of sporadic distribution in native waters may be correlated with success in culturing these larvae. Loosanoff and associates (Loosanoff, 1954, 1958) have reached the same conclusion. Our success in rearing clam larvae in North Carolina may be related to this phenomenon, and this in turn to the presence of moderate pollution in the sea water employed. We concur with Loosanoff that complete control of laboratory experimentation involving marine animals requires that these substances be characterized and further suggest that efforts be made to develop artificial sea water of known and reproducible composition for use in experimental study of marine animals.

STUDY OF INDIVIDUAL CLAMS. We studied the behavior, growth, and form in individual veligers and early post-set stages of *Mercenaria* in running-sea-water microculture dishes constructed as follows. A collar of plankton cloth (18XXX) 3 cm. high was fastened to a watch crystal (1 cm. high and 2.6 cm. in diameter) with clean melted paraffin (Fig. 3). Several dishes were placed around a 2.5-liter evaporating dish and each connected with this by means of siphons bent from glass tubing 3.5 mm. in diameter. The opening of each siphon in the reservoir ranged from 200 to 250 μ in diameter and in the microculture dish, 500 to 700 μ. When the bottom of a microculture dish was placed 1.5 cm. below that of the reservoir, each siphon delivered about one drop per second when the reservoir was full and about one drop in six seconds when the container was nearly empty. This rate of flow

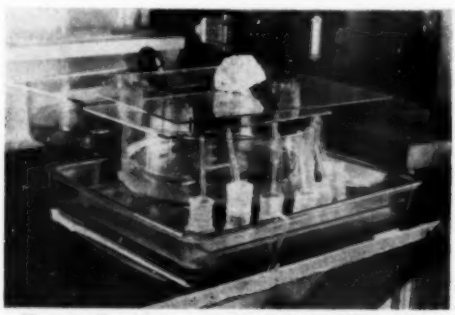

FIG. 3. Running sea water microculture dishes for culture of individual larval and early post-set *M. mercenaria*.

and treated with bone charcoal. Larvae grew most rapidly and with negligible mortality in (a); they started to set in eight days, and all larvae metamorphosed by the twentieth day, at which time the larger ones measured 0.55 mm. in length. In (b) setting commenced in eleven days, also with negligible mortality; and also in twenty days all larvae had set; the largest measured 0.33 mm. in length. In (c) and (d) larvae grew very slowly; an occasional larva started to set in thirteen days, and by twenty days only a few had fully set in (c) and very few in (d). Maximum lengths at the end of twenty days averaged 240 μ in (c) and 220 μ in (d); some mortality occurred in both cultures. The experiment suggests that laboratory running sea water (the intake pipe was heavily fouled with living sessile organisms at the time), heat-treated sea water, and charcoal-treated sea water each successively removed more of some "substance" from the water which is necessary for growth.

[3] At least part of our difficulty in raising clam larvae on Gardiners Island may have been the presence in many of our cultures of the parasitic fungus *Sirolpidium* (Davis et al., 1954), which we identified within numerous larvae during the summer of 1954. We encountered no such parasite in Morehead City, but a small copepod, *Tisbe* (= *Idya*) sp., a common co-inhabitant of many of our clam larval cultures which fed on the algae and reproduced actively, inhibited feeding to a slight degree by moving rapidly over newly set clams. The copepods entered cultures in sea water as young stages and then could not be removed mechanically because their size range corresponded with that of clam larvae. Loosanoff et al. (1957) have since developed a chemical control for these crustaceans.

produced a relatively constant gentle circulation in each microculture dish. Introducing additional reservoirs siphoning in series provided a system which operated unattended for twenty-four hours. Approximately 35 cc. of mixed algal food culture, screened through a No. 325 stainless steel screen, per liter of fresh sea water were added daily to the reservoirs. The flow of sea water through the plankton-cloth collar of each dish was maintained for about two weeks by occasional brushing of the cloth and removal of detritus accumulating within the dish with a small bulb pipette. When the plankton cloth fouled, *Mercenaria* were transferred to clean collared dishes by means of finely drawn glass bulb pipettes. After metamorphosis, larvae tended to remain on the bottom, and clogging of the plankton cloth and flow of water over the top of the collars was less likely to carry clams out. The addition of a thin layer of fine sand to the dish further reduced wandering of clams.

Thirty-five *M. mercenaria* ranging in length from 135 to 295 μ (these included both individuals cultured in the laboratory and some captured with the plankton net in native waters) and were isolated in microculture dishes. Maximum growth was obtained by a larva which was collected in Little Egg Harbor when it was 177 μ in length. It was maintained in captivity for 31 days and during this time grew to a length of 480 μ.

II FUNCTIONAL MORPHOLOGY

Definition of Early Stages of Life Cycle

During larval development and metamorphosis the young of *M. mercenaria* pass through a series of well-defined recognizable stages characterized by distinct structural features and behavior. Table II suggests a terminology for these stages which is based, when these are available and suitable, on terms already employed in the literature of bivalve larvae (Werner, 1939; Jørgensen, 1946; Rees, 1950; Quayle, 1952). Stages are defined principally on the basis of valves, locomotor organs, byssus, siphons, and the spatial position of the animal in its native habitat. Valve dimenions accompanying the appearance, and in some cases subsequent disappearance, of larval organs are only generally diagnostic of larval and post-setting stages, since shell dimensions of successive stages overlap broadly in different individuals. Thus length limits given in Table II are offered only as rough indicators of the stages, and those given for early post-set stages are incomplete. The position and behavior of young hard clams in the native habitat change with each successive major morphological development; thus identification of stages is useful in studies not only of functional morphology and physiology, but also of behavior and autecology.

Planktonic Stages

1. Nonshelled

When first discharged by the female, ova of *M. mercenaria* are slightly grayish and granular in appearance. They vary in diameter from about 60 to 85 μ and are surrounded by a conspicuous, clear, gelatinous envelope about 25 μ in thickness (see Figure 1, Loosanoff and Davis, 1950). The envelope soon swells and in four hours may attain a thickness of 95 μ, giving a large ovum an over-all diameter of approximately 270 μ. By the late gastrula stage, the embryo, still in the gelatinous envelope, whirls rapidly on its long axis. In 12 hours at room temperature the early trochophore, which is oval in shape, emerges from the envelope; it has a dense mat of long cilia over the equatorial region and a prominent single apical flagellum. How the embryo escapes from its gelatinous envelope has not been described. The embryo is now approximately 85 μ long and its apical flagellum equals the length of the embryo. It swims rapidly through the water, holding the flagellum straight in front of it; when held under a cover glass the embryo periodically whips the flagellum basally. In about 16 hours the trochophore assumes a spherical shape, and a broad area of long velar cilia, 7 μ in length, develops around the base of the apical flagellum; short cilia clothe the remainer of the embryo. Shortly thereafter, when it has length of about 90 μ, the embryo starts secretion of prodissoconch I valves, and at an age of 20 to 24 hours the fully shelled straight-hinged veliger appears (Figs. 4, 5). Appearance of the valves does not seem to decelerate swimming rate, possibly because the size and efficiency of the velum keep pace with the growing larva. (See Belding [1912] for a description of the general embryology of the early stages of the hard-clam larvae, which he could raise only to the straight-hinged stage.)

Table II

Summary definition of early stages of the hard clam, *Mercenaria mercenaria*, based on laboratory-reared animals.

A. PLANKTONIC STAGES:

1. NONSHELLED: development from fertilized ovum through blastula, gastrula, and trochophore to nonshelled veliger stage occurs in about 16 hours. Early stages float passively in water currents; trochophores and naked veligers actively propel themselves through the water by means of a strongly ciliated velum (Fig. 1, Loosanoff and Davis, 1950).
2. SHELLED (*prodissoconch*):
   a. *Straight-hinged veliger:* possesses smooth valves; velum continues to grow in size and larva is a strong swimmer; length range, 90-140 μ, age range, 1-3 days (Figs. 4, 5).
   b. *Umboned veliger:* smooth valves continue to grow symmetrically, but now a gently sloping umbone projects above middle of hinge line; length range, 140-220 μ, age range, 3-20 days (Figs. 5, 6).
   a. *Prodissoconch I* (not synonymous with straight-hinged veliger): first valves: thin, uniform, transparent, secreted by surface of mantle, appear during first 24 hours of life (Figs. 4, 5, 6).
   b. *Prodissoconch II* (Werner's *reliconcha stage*, not synonymous with umboned veliger): valves added to Prod. I by edge of mantle and mantle surface after first 24 hours of life during remainder of veliger stage, still smooth but with growth striae (Figs. 5, 6).

B. PEDIVELIGER (SWIMMING-CREEPING) STAGE:

At an age varying from 6 to 20 days and a length ranging from about 170 to 220 μ in different individuals, the veliger gains use of a foot, and thereafter alternates swimming in water and crawling on the bottom; the stage appears to endure for a variable period in different individuals and terminates at a shell length ranging from about 200 to 230 μ, when the velum is lost (Fig. 6).

C. PLANTIGRADE ("CRAWLER," DISSOCONCH) BENTHIC STAGES:

After the velum is lost and beginning at a length ranging from 200 to 230 μ in different individuals, locomotion in the metamorphosing larva is limited to crawling on the foot on the bottom:

1. BYSSAL PLANTIGRADE: soon after—and sometimes before—losing the velum, the plantigrade affixes to the substratum by means of a byssus (thus reaching the *setting*, or "spatting," stage); thereafter for a number of weeks (to a shell length of approximately 9 mm.) it alternates byssal attachment and active crawling, remaining on or superficially in the sediment; siphons form, and as they lengthen the clam moves deeper into the sediment; shell secretion now takes the form of concentrically ridged dissoconch valves, small primary ridges being followed by more conspicuous secondary ridges (Fig. 7).
2. JUVENILE PLANTIGRADE: young clams approximately 9 mm. and longer in which the byssus gland is no longer functional, the byssus is lost, and the animal maintains its position beneath the surface of the sediment by means of the foot alone; definitive siphons are fully developed (Fig. 12).

2. Shelled

STRAIGHT-HINGED VELIGER. The early straight-hinged veliger has a stippled gray appearance with a slight tinge of translucent grayish-salmon color which fades to clear at the edges of the valves. Stippling is uneven and most pronounced in the center. Globules occur throughout the protoplasm, range in color from clear to dark gray, occur in a loosely scattered pattern, and vary in size from 1 to 14 μ (Figs. 4, 5).

An early straight-hinged veliger of average dimensions, when first fully covered by valves, possesses a length (maximum anterior-posterior distance parallel to the hinge line) of 98 μ, a height (maximum distance between the hinge line and the ventral curvature of the valves) of 78 μ, a hinge line 65 μ long, and a width (maximum distance between external surface of the two valves) of 48 μ. Although the ratios of these dimensions remain relatively constant, the size of larvae varies considerably, lengths as small as 86 μ being recorded by Loosanoff et al. (1951b).

When held between a cover glass and a slide, straight-hinged larvae commonly swim against the surface film of air bubbles in a stationary position, actively beating the cilia of the velum. Under these circumstances it has been possible to obtain the dimensions of the velum and its parts. A larva 125 μ in length possessed a velum 65 μ in diameter, velar cilia about 20 μ in length, and an apical flagellum

FIG. 4. Detailed drawings of straight-hinged veligers of *M. mercenaria* reared in the laboratory. Left, 100 μ long, 250X; right, 120 μ long, 260X.

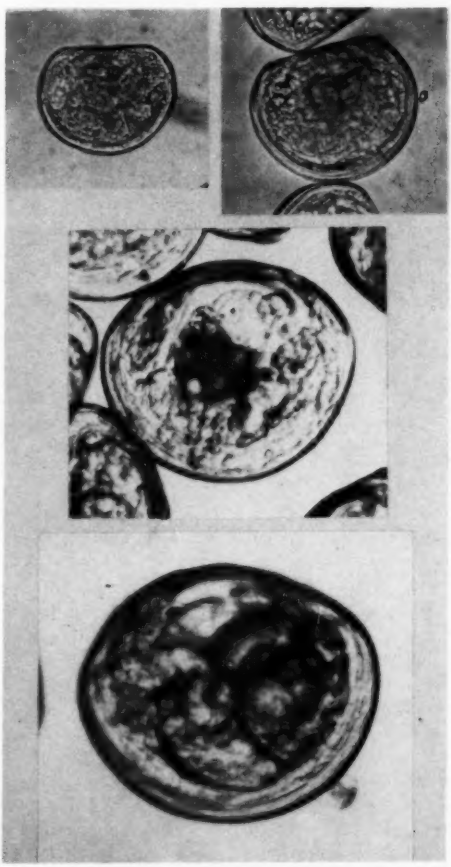

FIG. 5. Photomicrographs of veliger larvae of *M. mercenaria* reared in the laboratory. Top left, straight-hinged veliger 90 μ long, 230X; top right, straight-hinged veliger 115 μ long, 230X; middle, early umboned veliger 145 μ long, 270X; bottom, late umboned veliger 180 μ long, 260X (photomicrographs by T. C. Nelson).

35 μ in length. Viewed from the hinge line (a dorsal view) with valves fully opened and velum active, rapidly beating velar cilia resemble a vibrating halo as their tips project beyond the sides of the valves. Velum retractor muscles, leading from the region of the hinge line to the inner circular base of the velum, are visible in swimming larvae at a magnification of 100X.

Valves of straight-hinged veligers are asymmetric, one end being slightly pointed and the other rounded; the hinge line is relatively long in proportion to the length and height and is straight (Fig. 4). Occasionally empty veliger shells were collected in plankton samples and compared closely with those of larvae cultured in the laboratory. Valves of natives did not differ significantly in appearance from those reared in the laboratory.

Initial shell material, prodissoconch I, which covers the earliest straight-hinged veliger, is very thin, transparent, smooth, and fragile. This is the stage which Werner (1939) called the "veliger"; the D-shaped shells covering it he called "prodissoconch I." Werner's use of the term veliger, however, is too restricted, since the larva possesses the velum throughout its planktonic existence. The term prodissoconch I for the first valves has gained acceptance and is used here; but Werner's term veliger will be extended in this paper to include all stages possessing the velum (Table II).

UMBONED VELIGER. Shell deposited after prodissoconch I, and for the remainder of the planktonic veliger existence, is characterized by mintue growth striae, though the over-all aspect of the shell surface is smooth. This secondary larval shell Werner calls prodissoconch II, and the larval stage accompanying it he names, appropriately, the "veliconcha" stage.

Infrequently prodissoconch II valves were collected in the plankton in which the edge of a valve had been broken off accidentally and replaced by new shell, in much the same manner as adult clams renew fringes of valves lost through unsuccessful attempts of predators to penetrate them. At a length of about 120 μ a pronounced line in the form of a smooth arc appears just under the hinge line at a point where the umbones are forming. Under a magnification of about 430X, minute punctate markings may be seen over the nascent umbones (Fig. 4, right). These markings are not visible in the photomicrographs (Fig. 5). A narrow band of light yellow color appears in the shell material itself as a rim around the outer margins of the valves (prod. II). This color is present in empty valves, and therefore is not pigment in underlying tissues. The medial part of the

shell (prod. I) is colorless. Tips of the umbones in the late straight-hinged veliger, at a shell length of 130 μ, approach close to the hinge line. At a length of approximately 140 μ the larva, now known as an early umboned veliger, is characterized by umbones which rise slightly but uniformly above the hinge line (Fig. 5, middle), and the punctate markings on the external surface of the umbones become more conspicuous. The band of light yellow shell around the valve margins persists.

Between a total shell length of 140 to 200 μ, umbones in the veliger continue to rise gradually but inconspicuously above the hinge line, giving the larva a rounded appearance in lateral profile (Fig. 5, bottom). Umbones never assume the prominence represented by those of the late veliger stages of *Crassostrea*, *Teredo*, and *Anomia*. As the umboned veliger of *M. mercenaria* grows, its straight hinge is eclipsed, one end of the larva becomes slightly angled while the other remains rounded, and shoulders off the ends of the umbones slope gently. Light yellow shell margins persist even on a diet of various micro-organisms possessing different pigments.

Whereas most tissues of healthy living clam veligers are grayish in color, the digestive diverticulum is often brightly pigmented, the color varying somewhat with the quality and quantity of the diet. As also reported by Loosanoff et al. (1951b), we observed that actively feeding veligers cultured in the laboratory at one time or other exhibited digestive diverticula ranging in color from light yellow to orange-brown to bright orange to green, the color of the diverticula corresponding more or less with that of food organisms. Diverticula of native larvae collected in the plankton likewise varied in color. Starved larvae in the laboratory possessed noticeably less pigment. Loosanoff et al. (1951b) state that color of larvae of *M. mercenaria* is not a reliable diagnostic taxonomic character. It should be emphasized, however, that it is the color of the digestive diverticulum which is so variable; pigmentation of the valves themselves is more reliable.

Under a magnification of 150X freshly consumed food particles, averaging 1 to 2 μ in diameter, were seen actively rotating in little clusters in the stomach, propelled by active gastric cilia. Larger particles than this were not observed in veligers, suggesting that ordinarily larger food organisms may not be ingested.

Pediveliger Stage

Toward the end of its pelagic existence the veliger develops a strong, mobile, highly ciliated muscular foot and for an indefinite period alternates between a planktonic and benthic habit (Fig. 6). According to Thorson (1957)

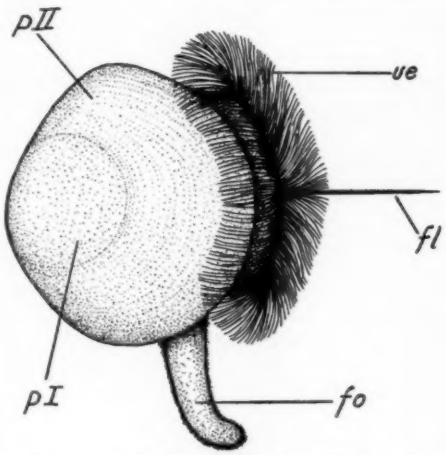

FIG. 6. Detailed drawing of pediveliger of *M. mercenaria* 200 μ in length, 210X. fl, apical flagellum; fo, ciliated foot; pI, prodissoconch I; pII, prodissoconch II; ve, ciliated velum.

the conjecture that larvae of many bottom invertebrates are able to respond positively or negatively to the substratum receives support from the "swimming-crawling stage," which seems to be a normal feature in development of lamellibranchs and marine gastropods, allowing them to crawl and carefully examine the substratum in which they have settled and then to swim up again. Because no term for this stage exists in the literature, and because of its ecological significance, the writer introduced the term "pediveliger" for it (Carriker, 1956 [misspelled "pedoveliger" in this publication]). Thorson (1946, 1950) suggests that probably most pelagic larvae have the capacity to postpone metamorphosis while seeking suitable substrata for the young, bottom-dwelling stage. In the hard clam, pediveliger "searching" (if indeed such occurs) is facilitated by the presence of both the velum and the foot. Whether metamorphosis in *M. mercenaria* is delayed in the absence of suitable substrata has not been determined. This should be investigated (Lynch, 1959).

At a length ranging from about 200 to 230 μ, and after the foot has developed, the pediveliger

loses the velum and takes up life on the bottom. The velum is not resorbed, but individual cells are shed and swim in the water for some time (Turner, personal communication). Raven (1958) speaks of the shedding of the velum in bivalve mollusks as a catastrophic event in which metamorphosis proceeds at first gradually and accelerates with a precipitate casting off of the whole velum in fragments. Cole (1938) reports from a careful study of the larval organs in the metamorphosis of *Ostrea edulis* that the bulk of the velar tissue is broken up and cast off or eaten, and the apical area of the larval velum forms the basis of the upper labial palps of the spat.

Loosanoff et al. (1951b) define metamorphosis in the hard clam as a "gradual replacement of a ciliated velum with a large muscular foot and ending with the development in the foot of a functional byssal gland." We infer that reference here is not to morphological displacement of the velum by the foot, since both organs are simultaneously functionally present in the pediveliger, or to the histological details of velar loss, but is rather a general statement of the process. Belding (1912) writes that the foot seems gradually to obliterate the velum, which can be seen in different stages of degeneration; the ciliated tip of the foot assumes the swimming function of the velum and the foot propels the clam through the water by a kicking movement. We are not able to confirm the functioning of the foot in this manner.

Internal anatomical changes accompanying metamorphosis have not been studied, and deserve attention. In this connection the study by Quayle (1952) of anatomical changes occurring during metamorphosis of the venerid bivalve *Venerupis pullastra* should be cited.

We concur with the observations of Loosanoff et al. (1951b) that hard-clam larvae obtained from a single pair of parents and grown under identical laboratory conditions often display considerable individual variation in the rate of growth and duration of the pelagic phase. These researchers found in their cultures that the maximum range of length of plantigrades at which setting occurred was from 175 to 236 μ, though it took place most commonly within a size range of from 200 to 210 μ. Larger larvae did not always set before smaller ones. Rate of growth of individuals within a single population varied widely: in some instances the length of larvae in a culture during the onset of setting ranged from 100 to 200 μ. Studying the effect of temperature on the rate of development of larvae, Loosanoff and associates found that at a constant temperature of 18°C. setting began 16 days and terminated 24 days after fertilization, and that at 30°C. setting began 7 days and ended 16 days after fertilization of ova. In general, within the temperature range of from 18° to 30°C. rate of growth increased with a rise in temperature. Loosanoff (1959) also discovered that the shape and size of metamorphosing larvae is not altered by development in different temperatures, a fact of some significance when the size and shape of prodissoconch II are employed in identification of the larvae. Landers (1954), in a three-year field study of the seasonal abundance of clam larvae in Narragansett Bay, Rhode Island, found that length of larvae of *M. mercenaria* in his samples ranged from 200 to 225 μ, when they disappeared from the plankton at the time of setting.

Plantigrade Stage

DISSOCONCH VALVES. With the genesis of dissoconch valve secretion by the outer surface of the outer fold of the mantle margin, distinct lamellate ridges appear on the external surface of the valves of byssal plantigrades (Fig. 12a–f). These consist of thin, more or less periodic elevations of shell surface which are concentric with the shell margin and run entirely around the valves from the hinge line. They were first observed by Belding (1912). Earliest ridges (here designated primary shell ridges), which are coincident with early life of the newly set byssal plantigrade on the surface of the sediment, are small and set close together and consist of soft, chalky-white shell material (Fig. 7 top, 12b–e). On the average, in faster-growing plantigrades the first ridge appears at a shell length of about 250 μ (Fig. 12a). In all, some four to twelve of these form on different individuals, but considerable variation in the number is apparent for a given size of clam. What determines the final number of these ridges is unknown. Because of the soft composition of the primary ridges, they soon erode, making them difficult to count on individuals under 10 mm. in length except on well-preserved specimens.

The deposition of a second set of lamellations (the secondary shell ridges) begins at a shell length of about one millimeter and coincides with early stages of development of the definitive exhalant siphon (Fig. 7 top, 10, 12d–e). These ridges are set more widely apart and are many times higher and thicker than the primary ridges; they are characterized by a translucent white color and are secreted periodically for the

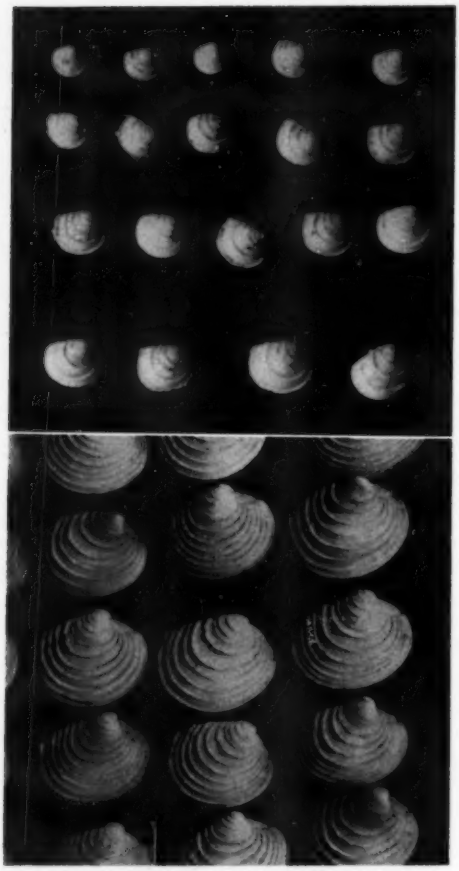

FIG. 7. Top, photomicrograph of byssal plantigrades of *M. mercenaria* ranging in length from 1.7 to 3.2 mm., illustrating the low, close-set primary shell ridges and the first two to five prominent, widely set secondary shell ridges; 3X (photomicrograph by T. C. Nelson and H. H. Haskin). Bottom, photomicrograph of juvenile plantigrades of *M. mercenaria* ranging in length from 11 to 13 mm., 1.5X (photomicrograph by T. C. Nelson).

duration of shell formation in the species. Secondary ridges are considerably harder than primary ones and consequently do not erode so readily.

In the summer of 1949 we made a study of the number and rate of formation of secondary shell ridges on the valves of byssal plantigrades. Fifty native young were collected locally in Little Egg Harbor, and fifty laboratory-reared hard clams were contributed by Dr. Loosanoff. Individuals of both populations were placed in sediment in small glass dishes in a protected enclosure secured to the side of the "Cynthia," which was moored in Thompson Creek, and examined periodically through the summer. When first collected and established in the enclosure the clams ranged in length from one to four millimeters. The number of secondary ridges on plantigrades of various lengths observed at intervals during the summer is given in Figure 8. There is a general tendency for larger clams to possess more ridges, but the number of ridges on different individuals of the same size is exceedingly variable. For example, the number of ridges in individuals in the length bracket 3.0 to 3.5 mm. varied from four to twelve; and a total of six ridges was found in a sample of individuals ranging in length from 1.0 to 4.6 mm. The number of ridges borne by laboratory-reared and native populations did not vary significantly.

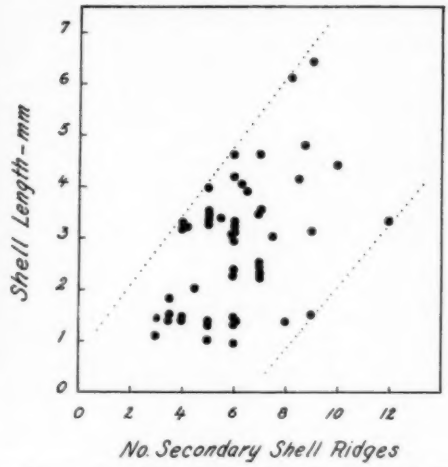

FIG. 8. Correlation of number of primary shell ridges with shell length in byssal plantigrades of *M. mercenaria.*

FOOT AND LOCOMOTION. The foot develops in the late veliger stage as a long, rounded, slender, heavily ciliated, highly supple, extensible organ (Fig. 6). We have seen functional feet in pediveligers as small as 166 μ in length. In clams 235 μ in length the foot may be extended normally about 190 μ beyond the ventral margins of the valves; this extension may be about 350 μ beyond the valves in individuals 415 μ in length, or 9 mm. in specimens 7 mm. in length (Figs. 9, 10). Inside the mantle cavity the foot may be turned 180° and protruded rearward, and after the foot contacts the substratum the body realigns itself over the

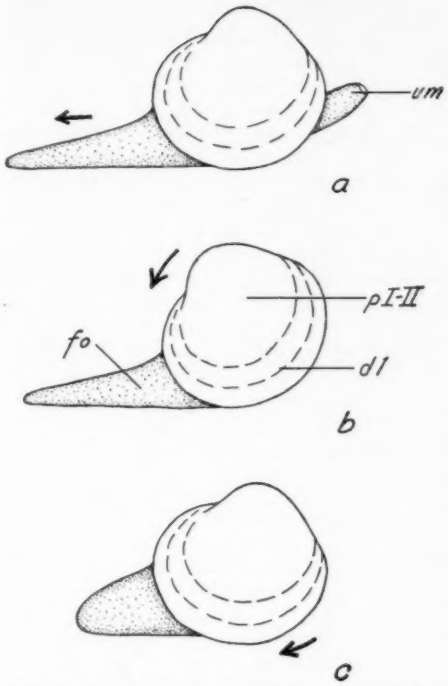

FIG. 9. Illustration of the mode of locomotion in a byssal plantigrade of *M. mercenaria*, 415 μ long, 80X. d1, dissoconch primary shell ridges; fo, foot; pI and pII, prodissoconchs I and II; vm, valvular membrane.

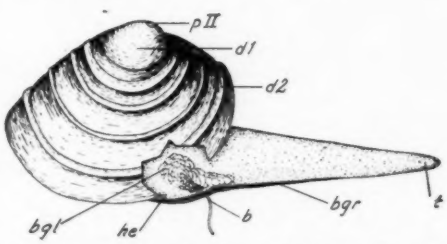

FIG. 10. Detailed drawing of byssal plantigrade of *M. mercenaria*, 4 mm. long, 10X. Portion of left valve cut away to show position of byssal gland in heel of foot. b, byssus; bgl, byssal gland; bgr, byssal groove; d1, dissoconch primary shell ridges; d2, dissoconch secondary shell ridges; pII, prodissoconch II; t, foot.

foot. On an open surface lacking sediment, clams often extend the foot under themselves and turn themselves over. As the plantigrade animal grows, the foot becomes shorter in proportion to the size of the shell, and the base of the foot becomes stockier.

Pedal locomotion in pediveligers and newly set plantigrades principally involves ciliary activity: cilia on the ventral surface of the foot beat toward the heel of the foot, so that the foot and animal glide forward smoothly over the substratum. Similar locomotion has been described by Quayle (1952) for planktonic veligers of *Venerupis* which have gained use of a foot. This type of movement is soon superseded in part by a muscular, crawling type of locomotion which is particularly well suited to movement through sediments and which is retained for the remainder of the animal's life. Crawling by byssal plantigrades about 0.4 mm. in length on hard, open surfaces is characteristic of all sizes of animals in this stage and agrees generally with the description given by Belding (1912) for crawling by *M. mercenaria* 2 to 3 mm. in length. It is performed in the following manner: from the resting position on one valve, a clam extends the foot anteriorly between the margins of the valves and effects contact with the substratum, the forward tip, or toe, of the foot acting as a searching and guiding organ (Fig. 9a). The mid-portion of the foot is then appressed to the substratum, flattened to one and a half times its normal width; the area of contact passes distad until the whole distal half of the foot is appressed and purchase with the substratum is effected. During this maneuver the clam draws its valves upright and pulls them forward over the foot by contraction of pedal muscles. If the foot, when first extended, strikes an object, it withdraws again within the mantle cavity and extends anew in a different direction, moving by means of ciliary and muscular-vascular action. Occasionally the valves fall again to one side but are soon righted. With each forward prolongation of the foot, the clam opens its valves slightly, and the mantle margins of the pedal opening maintain a close seal around the foot to exclude sedimentary grains as the foot is withdrawn; it simultaneously everts the exhalant valvular membrane (Figs. 9a, 12b–d). Upon initial contraction of pedal muscles, the valves close gently upon the foot and the valvular membrane is withdrawn completely (Fig. 9b); anterior portions of the valves are pulled toward the substratum (Fig. 9b), and immediately following this action ventral portions of the shell are drawn forward (Fig. 9c) over the foot as the foot contracts. The over-all effect is that of a bivalve moving forward by rocking its shells over the foot, the ventral rims of the valves sliding over the bottom much as a sled moves

on its runners. The foot adheres to firm substrates with great tenacity.

Clams express much the same type of locomotory activity when crawling through sediments. A pointed toe (Fig. 10) extends into the sediment and then expands, forming a partial anchor; then the foot contracts and draws the body over it. Clams are capable of pushing aside, or crawling under, grains of sediment several times the size of their bodies. Digging movements in young *M. mercenaria* resemble those described for young *Venerupis pullastra* by Quayle (1952), except for protrusion and use of the heel of the foot in digging by *V. pullastra*.

BYSSUS AND BYSSAL ATTACHMENT. Belding (1912) was the first to report that young *M. mercenaria* possess a byssus. Shortly after larvae of *M. mercenaria* assume the plantigrade habit, and sometimes before the velum is lost, the byssus gland becomes functional and the bivalve begins attachment to substrata by means of a transparent, somewhat elastic thread, the byssus (Feen, 1949). The byssus gland occupies a major portion of the heel of the foot (Fig. 10). Fluid byssal material passes from the gland through the byssal duct to the midventral base of the foot, where it flows into the byssal groove. This is a longitudinal depression along the midventral surface of the basal two-thirds of the foot. The groove shallows as it passes distally from the heel, and its edges, which form prominent folds in the vicinity of the heel, also diminish in size distally. Kellogg (1892) illustrates and describes the histology of the byssal gland in the carditid bivalve *Venericardia borealis*. The secreting epithelium is deeply folded, and at the inner ends of the folds many muscle fibers are strongly inserted and serve by their attachment to the valves dorsally as a powerful support for the byssus. No such studies have been reported for *M. mercenaria*. Young hard clams effect initial fixation to the substratum or to grains of sediment by means of a very fine transparent byssus scarcely visible at a magnification of 70X. As the clam grows the byssal gland, duct, and groove continue to develop. In individuals 270 μ in length the byssus possesses a maximum diameter of 5 μ and a length of about 0.3 mm., though the length is extremely variable; in an individual 400 μ in length, the byssus is 8 μ in diameter; in one 6 mm. in length, 30 μ in diameter and up to 4 mm. long to the first holdfast, and may extend through a number of holdfasts to a total length of 2 cm. (Fig. 11). By the time a clam reaches a shell length of 0.5 mm., the byssus is a sturdy anchor line; and at shell lengths ranging from 0.5 to 1 mm., clams adhere so firmly to hard surfaces by means of the byssus that a strong stream of water directed at them from a bulb pipette does not dislodge them. The longest byssus encountered proportionate to the length of the shell was 5.5 mm. in a clam 1.4 mm. long. We were not able to determine the range of lengths at which clams lose use of the byssal gland; a gross estimate of the average length is about 7 mm. Belding (1912) reports finding a byssus in a clam 9 mm. long.

We made the following observations of byssus formation and attachment: A binocular microscope was secured in an inverted position on a cantilevered support under a small petri dish lighted from beneath by a strong spot light. Clams ranging in length from 0.5 to 7 mm. were placed in shallow dishes with a small quantity of fine beach sand and clean sea water. Light did not appear to disturb the mollusks so long as they remained buried in the sediment. This device permitted the observer, lying in a steeply angled swivel chair under the microscope, to follow movements of plantigrades at various magnifications as they crawled about in the dish and effected byssal attachment. Observations were made in both New Jersey and in North Carolina on individuals of several populations of active, fast-growing, laboratory-reared *M. mercenaria*.

Clams anchored the byssus on an open surface but made attachment more quickly and frequently when completely buried under sediment. (We used the vital stain neutral red in preliminary observations to demonstrate the byssus more clearly, but the stain tended to inhibit attachment and therefore was discarded.) In the process of attachment under these laboratory conditions a clam extends the foot slowly, moving the foot through the sand with the toe held close against the substratum. At full extension of the foot the toe is pressed firmly against the substratum for a period varying from 0.5 to 3 seconds. During this action, edges of the byssal groove meet midventrally, converting the byssal groove into a tube, and minute peristaltic waves pass along the foot, this probably presses byssal fluid along the byssal tube, since formation and fixation of the byssus is too rapid to be effected by ciliary action alone. After attachment of the byssus to the substratum, the foot is withdrawn slowly (at the same rate as when crawling) within the mantle cavity, paying the byssus out behind

it. The actual attachment, or holdfast, of the byssus on the substratum is a hardened drop of byssal fluid. Probably upon contact with sea water, this hardens quickly, as indicated by the fact that the byssus holds against a stream of sea water from a small pipette within less than a minute after attachment. Following affixation, the clam may withdraw the foot completely within the mantle chamber and close the valves upon the byssus; or it may remain open, siphoning interstitial sea water under the sediment; or it may crawl to the surface of the sediment to siphon, still held by the byssus; or it may secrete a longer byssus with a number of holdfasts (Fig. 11) before becoming quiescent.

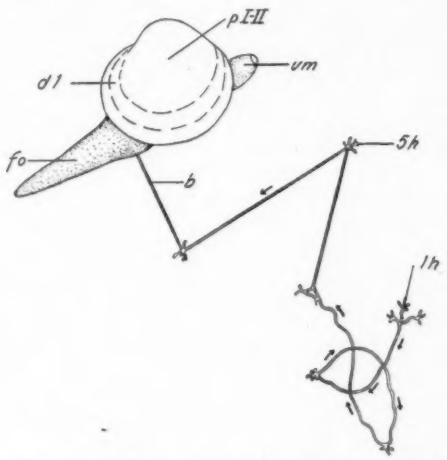

FIG. 11. Diagram of the byssal "trail" laid down by a byssal plantigrade of *M. mercenaria*, 415 μ long, in shallow sand in a finger bowl. 50X. b, byssus; d1, dissoconch primary shell ridges; fo, foot; pI-II, prodissoconchs I and II; vm, valvular membrane; 1h, first holdfast; 5h, fifth holdfast of byssus.

The holdfast is usually characterized by a distinct digitate blob of byssal material, frequently containing sand grains trapped in the byssal fluid as the holdfast is molded against a sand-covered firm surface. The small size of the holdfast (Fig. 11) suggests the degree of confinement afforded the hardening byssal fluid by the pedal tip at the moment of attachment. The complexity of the sample byssal trail illustrated in Figure 11 directs attention to the considerable versatility and maneuverability of the byssal groove and foot during placement of successive holdfasts.

In deep sediment clams affix the byssus to several grains of sediment a short distance under the surface and then move surfaceward, assuming a position, depending upon the length of the siphons, on or just under the water-sediment interface. In a dense population of byssal plantigrades, byssuses form a loose mat of interwoven threads.

Clams may also affix by means of a forked byssus, though actual formation of this was not observed.

Removal of sediment from about byssal plantigrades causes them to release the byssus at its insertion in the byssal duct, move to another site, and there reaffix. In discarding the byssus, young plantigrades crawl forward effortlessly, easily dropping the byssus and holdfast behind. Those approaching a shell length of 300 μ may make two or three attempts, circling about, resting occasionally, pulling tightly against the holdfast, until the byssus is dislodged from the byssal duct. A clam two millimeters in the branching ducts of the byssal gland it-at the holdfast for a minute or so before crawling away. One such individual, in detaching the byssus, crawled slowly and with apparent effort from the holdfast over a period of 5 minutes, stretching the byssus to a length of 5.5 mm. before it gave way, contracting onto the substratum like a rubber band. Whether the byssus is held in the duct by a sphincter muscle or by hardening of the byssal branches in length may turn about on its foot and tug self is not known. Probably the closed valves further aid in securing the byssus to the animal.

In the laboratory the interval between disturbance and reattachment of different individual byssal plantigrades varied from a few minutes to several hours. We made this observation in North Carolina in August, 1956, on laboratory-reared clams ranging in length from 0.9 to 2.4 mm. These animals were maintained in running, sand-filtered, sea water and fed a supplemental diet of mixed algae. Twenty clams were placed in each of 10 petri dishes 5 cm. in diameter with a little heap of fine beach sand and fresh sand-filtered sea water (salinity 35.5 °/oo). Clams and sand were swirled into the center of each dish and the dishes set aside under glass covers. No artificial aeration or food was provided during the observations. Room temperature during this period varied from 23°C. in the morning to 30°C. in the late afternoon. At varying intervals of time (Table III), clams which had affixed were exposed by gentle flushing aside of the sand in each dish with a stream of sea water from a bulb pipette. The rate of attachment is recorded in Table III. In a

Table III

Rate of byssal attachment by *M. mercenaria* in the laboratory.

Interval clams undisturbed in sand, mins.	Percentage of clams attaching during this interval
5	5
10	7
15	17
20	19
30	48
40	32
50	42
60	39
120	32
180	60

second experiment under similar conditions of salinity and water temperature, 150 clams similar to those employed in the experiment reported in Table III went uninterrupted for eighteen hours. Ninety-one per cent of these made byssal attachment, indicating that in a longer period, most of the clams did attach. Under other less carefully controlled conditions on Gardiners Island attachment of laboratory-reared clams took place more rapidly, a reaction probably more accurately representing that in nature.

Byssal attachment is not an uncommon means by which young bivalves anchor themselves to the substratum and thereby remain closer to the general area of setting (*Mytilus edulis*, Kellogg, 1892; *Mya arenaria*, Kellogg, 1901; *Hiatella gallicana*, Hunter, 1949; *Venerupis pullastra*, Quayle, 1952; *Lasaea rubra*, Morton, 1960). This is the first description of the process in *M. mercenaria*.

MANTLE FUSION AND SIPHONS. In newly metamorphosed asiphonate individuals of *M. mercenaria*, margins of the mantle lobes are free, and they part as the valves gape to permit both movement of sea water through the mantle cavity and extrusion of the foot. At a shell length varying in different individuals from about 210 to 300 μ, the inner folds of the posterioventral portions of the margins of the mantle lobes fuse over a short distance; this forms a tissue union, the siphonal septum, which divides the over-all gape into an exhalant and a combination pedal-inhalant opening (Fig. 12a). The latter functions in the intake of water and as an opening for pedal activity. We are basing anatomical terminology for *M. mercenaria* on that developed by Yonge (1957) in his study of mantle fusion in lamellibranchs. From the rim of the exhalant aperture thus formed, there soon emerges a very thin, transparent, pliable, tapering sleevelike tube, the valvular membrane (Fig. 12b), which is an extension of the inner folds of the mantle lobes and controls and directs the exhalant current. Minute particles passing out of the mantle cavity through the valvular membrane, which now accommodates the excurrent flow of sea water from the mantle cavity, are easily visible through the walls of the membrane. As the clam opens its valves to pump sea water, the membrane billows outward rapidly, forced by pressure created within the mantle cavity through ciliary activity, the distal tip unfurling from within. When hydrostatic pressure is momentarily cut off by constriction of the inhalant siphonal membrane (also see Sellmer, 1959) across the internal base of the siphon, or by the meeting of the mantle margins with no change in volume of the mantle cavity, the valvular membrane collapses into a shrunken, wrinkled tube which dangles externally. More frequently, the membrane when not functioning is inverted swiftly within the mantle cavity, distal tip first, possibly as a result of the apposition of the mantle edges concurrently with slight enlargement of the mantle cavity. Belding (1912) briefly describes and illustrates the membrane in young *M. mercenaria* and was probably the first to report it in this species.

During growth of byssal plantigrades to a shell length of about 1.5 mm., the valvular membrane continues to enlarge (Table IV), though the ratio of shell length to length of membrane in different individuals of the same size is quite variable. Thereafter, between shell lengths of approximately 7 to 20 mm., dimensions of the membrane decrease in proportion to the size of the animal as the animal grows. Eventually the membrane is reduced to a shallow flange at the tip of the definitive exhalant siphon just inside the circle of siphonal tentacles (Fig. 12b–g). Particularly during its more prominent stages, the valvular membrane is characterized by flexibility and by a relatively small distal orifice which reduces the diameter of the exhalant siphon to a fraction of that of the inhalant siphon (Fig. 12e–g).

Shortly after the genesis of the exhalant valvular membrane, two small truncated tentacles, forerunners of the ring of sensory ten-

Table IV

Ratio of length of shell to length of valvular membrane in ten young plantigrade *M. mercenaria* selected at random for measurement.

Shell length, μ	Length of valvular membrane, μ	Ratio of length of shell to length of valvular membrane
230	50	4.6
290	—	—
330	100	3.3
415	200	2.1
625	380	1.6
1000	780	1.3
1360	615	2.2
1500	1500	1.0
4000	1420	2.8
14000	1100	12.7

tacles which will adorn the distal rim of the inhalant siphon, appear anteroventrad the base of the valvular membrane (Fig. 12b). Then two more tentacles form anterior to the first pair. Simultaneously the anterior portion of the fused mantle splits at the site of the inhalant siphonal opening (Fig. 12c); this makes possible conjoining of inhalant and exhalant siphonal walls as they develop. At this time a single tentacle appears dorsal to the base of the valvular membrane, the first of a ring of tentacles which will bound the distal opening of the definitive exhalant siphon. Tentacles continue to form anteroventrad the site of the first ones as definitive inhalant and exhalant siphons develop (Fig. 12d). By the time the two siphons are visible beyond the margins of the shells, inhalant and exhalant tentacles from complete rings on the distal orifices of the siphons (Fig. 12e), and the valvular membrane, now projecting from the inside base of the border of tentacles, is borne on the distal end of the lengthening exhalant siphon.

Before the appearance of the inhalant siphon, water is drawn into the mantle cavity through the inhalant-pedal opening, and the clam remains at the sediment-water interface with mantle margins in contact with ambient sea water. As siphons form, the animal begins to burrow shallowly in the sediment, siphons upward. It moves deeper as siphons lengthen, and eventually the entire shell disappears within the sediment. Direct contact with overlying sea water is maintained by siphons extending to or shortly above the surface of the sediment; or contact may be indirect through interstitial water when siphons are held beneath the sediment. Yonge notes (1957) that formation of siphons was a matter of major significance in the evolution of bivalves, enabling them to live in the shelter of a soft substratum. Clearly this step in its evolution has permitted *M. mercenaria* to survive abundantly in spite of its numerous predators.

In *M. mercenaria* the siphons are of type B, according to Yonge's (1957) classification, in which the combined siphons are formed as extensions of inner and middle folds of the mantle margins. Like the valvular membranes, siphonal tentacles are also derived from the inner fold of the mantle margins. Mantle fusion and siphon formation in *M. mercenaria* resemble rather closely that described briefly by Quayle (1952) for the venerid clam *Venerupis pullastra*.

Arrows in Figures 12a–g indicate direction of flow of sea water through the mantle cavity. In asiphonate byssal plantigrades restricted to lying or crawling on the surface of sediments, the ventral margins of the mantle lobes separate slightly to permit ingress and egress of sea water, the region of inflow occupying more than half of the length of the gape. Initial fusion of the mantles at the siphonal septum restricts egress of water to the posterior or exhalant opening. As the inhalant siphon forms, it becomes the principal course of entry of incurrent water into the mantle cavity, and the pedal gape is utilized only occasionally.

In siphonate clams, siphon tips (including the valvular membrane) point away from each other (see especially Fig. 12f–g), thus deflecting incurrent from excurrent water. As demonstrated in hard clams 10 to 15 mm. in length in sea water containing a carmine suspension, the excurrent stream, which flows from the relatively small exhalant orifice, is characterized by a velocity and length of flow path several times that of the incurrent flow through the larger inhalant opening. As a consequence of this differential in flow, the water mass in the immediate vicinity of the siphons is moved away from the posterior end of the clam, drawn by the relatively stronger flow of the excurrent stream, and mixing of incoming and effluent water is thus much reduced. Therefore, in moving water a siphonate clam draws in little, if any, of its own excurrent water; and in still water some time may elapse before excurrent water recirculates to the inhalant siphonal aperture. The valvular membrane functions provisionally

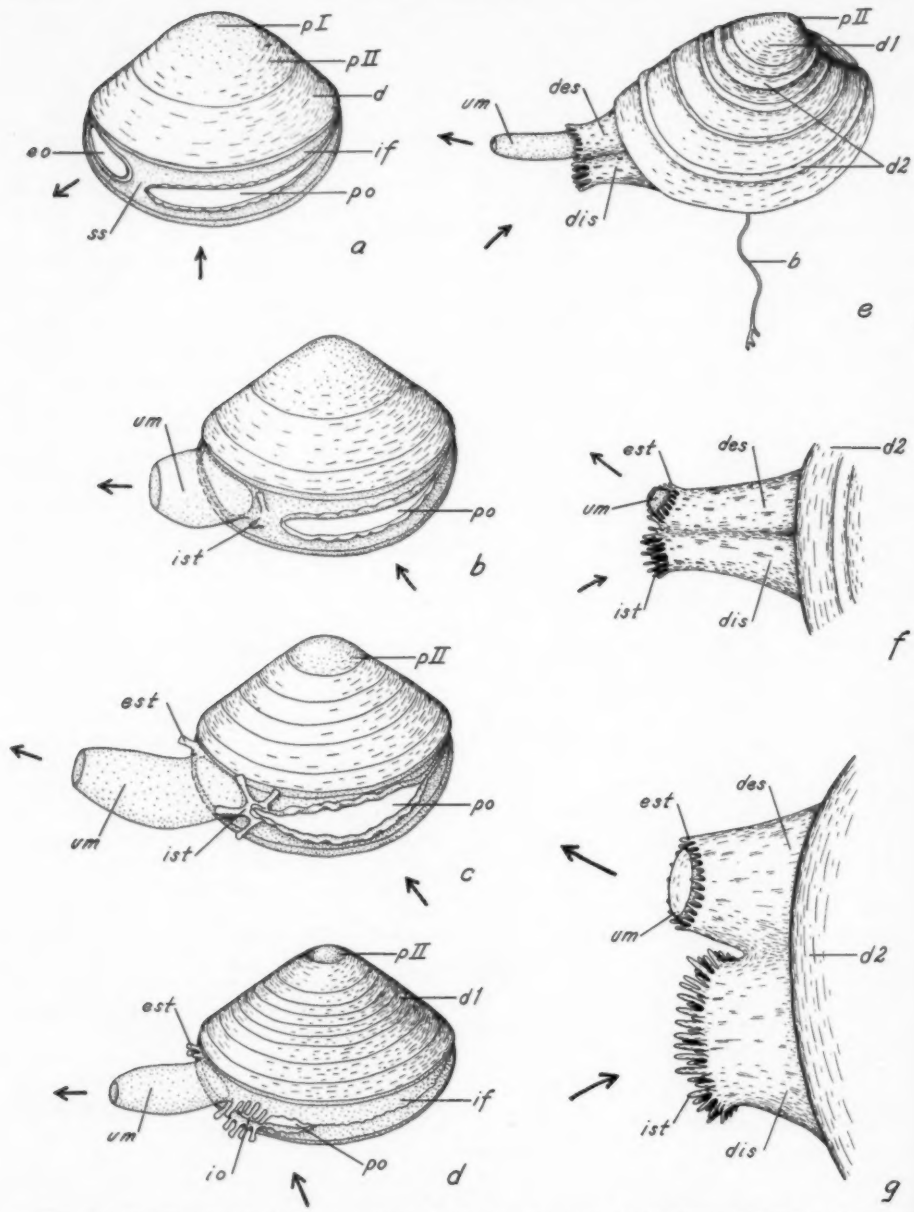

FIG. 12. Detailed drawings of seven stages in the development of definitive siphons in *M. mercenaria;* arrows indicate direction of flow of water through clams:

a. Byssal plantigrade, 290 μ long, 250X. Fusion of mantle margins delimiting opening for the exhalant siphon has begun; deposition of dissoconch shell has started, and two primary shell ridges are present.

b. Byssal plantigrade, 330 μ long, 220X. Valvular membrane is forming out of exhalant opening, and two inhalant siphonal tentacles and two primary shell ridges are present.

c. Byssal plantigrade, 625 μ long, 115X. Valvular membrane is further developed; four in-

in deflecting excurrent and incurrent streams during the period of definitive siphon formation. Quayle (1952) assigns to the valvular membrane in young sedentary *Venerupis* the function of keeping the mantle cavity and gills free of feces. Newly metamorphosed *M. mercenaria* undergo a similar sedentary asiphonate stage in which, so far as can be determined, however, no fouling of surfaces of the mantle chamber occurs. Thus it would appear that in *M. mercenaria*, at least, the valvular membrane serves as a transitory exhalant siphon, insuring a decreasing rate of recirculation of mantle sea water as the clam grows and as the area of its respiratory membranes decreases relative to the increase in body volume and greater need for oxygen.

ROLE OF VALVES, FOOT, AND SIPHONS IN BURROWING. The activity of byssal plantigrades within sediments was observed under low magnification in small sediment-filled glass cells. Open rectangular receptacles which were constructed of glass plates and were slightly wider internally than the width of young clams were filled with fine, muddy sand and sea water and secured openside up against the stage of a binocular dissecting microscope held in a horizontal position. Clams, confined loosely between the sides of the cells, moved up and down readily and could be followed closely through the microscope, illuminated by direct light from a laboratory window. Sea water in the cells was changed frequently. Some clams were dropped on the surface of the sediment and observed as they buried themselves, and others were placed deep in sediment and watched as they moved surfaceward. Observations were made principally on Gardiners Island in prevailing summer salinities and temperatures there.

In burrowing, the clam extends the foot downward out of the mantle cavity, lengthening it to a long, slender, tapering organ which readily worms its way among the grains of sediment. At the maximum extension of the foot the distal tip distends, providing a partial anchor in the sediment against which the remainder of the foot pulls as it contracts and draws the anterior end of the clam downward into the sediment. Immediately after, the clam draws its posterior end downward and forward, the direction of movement of the valves more or less paralleling the direction of the concentric shell ridges. In this manner these ridges offer minimum resistance to progress of the valves through sediment; yet after the clam becomes stationary, the ridges, because of their size and curvature, themselves aid in anchoring the whole animal in the sediment. The seesaw movement of the valves continues until the clam has completely buried itself. In loose sediment a byssal plantigrade can bury itself in a half-dozen seesaws in less than a minute.

When coming to the surface a byssal plantigrade forces itself upward out of the sediment with the foot; it moves slowly and intermittently, more or less posterior end first, and omits the seesaw movements of the valves. If siphons point in some other direction, the clam reorients itself upside down. The foot extends slowly, pressed firmly against the sediment; it withdraws to variable lengths, rests for several seconds, and then extends anew to push the body further up. The entire maneuver is an un-

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halant siphonal tentacles, one exhalant siphonal tentacle, and five primary shell ridges are present.

d. Byssal plantigrade, 1.36 mm. long, 55X. Valvular membrane is further enlarged; ten inhalant siphonal tentacles, three exhalant siphonal tentacles, and ten primary shell ridges are present.

e. Byssal plantigrade, 4 mm. long, 20X. The valvular membrane has been carried outward on the distal end of the definitive exhalant siphon as the latter developed, and the inhalant siphon has formed alongside and fused to the exhalant siphon. Numerous tentacles are present on the distal margins of both siphons. The byssus extends ventrad between the valves, and four secondary shell ridges are present. Prodissoconch and dissoconch valves with primary shell ridges are illustrated diagrammatically as smooth shell surface.

f. Siphons of juvenile plantigrade 14 mm. long, 4X. Siphons are fully formed, and the valvular membrane has been reduced to a short, funnel-shaped outlet.

g. Definitive siphons of adult, 55 mm. long, 4X. Siphons extend only slightly beyond the margin of the valve, though they many be extended very much farther (see Fig. 13), and the valvular membrane is now reduced to a short shelf just inside the tentacular border of the exhalant siphon.

b, byssus; d, dissoconch; des, definitive exhalant siphon; dis, definitive inhalant siphon; d1, dissoconch with primary shell ridges; d2, dissoconch with secondary shell ridges; eo, exhalant opening; est, exhalant siphonal tentacle; if, inner fold of mantle; io, inhalant opening; ist, inhalant siphonal tentacles; po, pedal opening; pI, prodissoconch I; pII, prodissoconch II; ss, siphonal septum; vm, valvular membrane.

hurried one, and there is no indication of distress. Byssal plantigrades can emerge from sediment through a depth of at least five times the shell length, and adult clams buried by sediment from human channel-dredging activities can move upward through at least a foot of loose, sandy mud at summer temperatures.

When crawling in sediment, the byssal plantigrade continues to pump water, though at a reduced rate which apparently corresponds with the porosity of surrounding sediment. Tentacles on the inhalant siphon fold inward, forming a partial screen which admits water and small particles but excludes the larger sedimentary grains. As the screen becomes blocked by larger particles, the clam appresses its valves slightly but forcefully, emitting spurts of water principally out the inhalant siphon. The force of these jets of water is considerable. For example, a clam three cm. in length buried in sediment with siphons extended just out of shallow water can jet a stream of water across an aerial trajectory some 30 inches from the inhalant siphon. In clams submerged in sediment, these jets clean the inhalant siphonal screen, and they also tend to clear a small sediment-free space in front of both siphons into which the siphons can then distend. The exhalant valvular membrane and siphon do not function in clearing a path for the clam through the sediment by discharging water, but in a limited way as permitted by confinement of surrounding sediment they do divert incurrent from excurrent siphonal streams of water. As the clam approaches the surface, siphons many extend to the surface and siphon actively, or may pump interstitial water just beneath the surface of the sediment. In Delaware Bay, New Jersey, Dr. T. C. Nelson (personal communication) occasionally observed only single excurrent streams of sea water bubbling from young clams in the sand, indicating that incurrent water was being drawn interstitially through the sediment. More commonly both siphons communicate directly with water at the surface.

Clams can extend the siphons beyond the margin of the valves to a length at least twice that of the valves. A series of measurements of siphon lengths in clams ranging in length from 5 to 35 mm. under conditions of low oxygen tension in trays and in jars (Fig. 13) established a range of ratios of length of siphon to length of shell of 1.0 to 2.3. The degree to which siphons may be extended normally in sediment in the native habitat is not known. Because of the capacity of *M. mercenaria* to

FIG. 13. Definitive siphons of adult *M. mercenaria*, shells ranging in length from 5 to 6 cm., and siphons fully extended in deep jar under conditions of low oxygen tension. 1/3X.

siphon within porous sediments, short siphons do not impose a limit on the depth to which the animal can burrow. In the field in the summer we have found young clams buried in sediment as much as five times the shell length; and T. C. Nelson (personal communication) reported finding the ventral margins of adult clams eight to ten inches below the surface of the botton in Delaware Bay in late November winter conditions.

Thus in nature young plantigrade *M. mercenaria* are well endowed for an existence of vertical adjustment to fluctuating bottom levels in unstable bottoms of channels and inlets where they commonly occur in abundance. It is probable that young byssal plantigrades cannot burrow until the ring of tentacles is available on the inhalant siphon as a screen to exclude sedimentary grains. Whether a change in geotactic response occurs at this time is not known.

III BEHAVIOR AND AUTECOLOGY

Planktonic Stages in the Laboratory

Trochophore and early straight-hinged stages of *M. mercenaria* are strong swimmers and tend to concentrate in the surface layers of water in culture jars. Turner and George (1955) determined that early straight-hinged veligers, when held at a temperature of 22°C., swam upward at a rate of from seven to eight centimeters per minute, showed no detectable reaction to light, and were indifferent to a considerable range of salinity gradients. These investigators introduced larvae into the bottom of a glass tube in which layers of diluted sea water were placed one above the other. The larvae passed through the sharp transitions separating layers which differed by as much as 5 °/oo with no loss in velocity until the gradient between 20 and 15 °/oo was traversed. At the latter stratum swimming velocity decreased and larvae swam slowly in circular patterns just above the 20–15 °/oo interface.

Late straight-hinged and early umboned veligers continue to be vigorous swimmers and tend to remain uniformly distributed in culture jars. They swim in slow, circling movements propelled by the ciliated velum projecting between gaping valves. When pipetted into a watch glass, some veligers remain on the bottom with valves closed momentarily while others swim immediately and actively about, a few so swiftly that they are difficult to follow under a magnification of 35X. In swimming off the bottom, some individuals spread the valves, extend the velum quickly, and swim hinge line first in spiraling paths; then they may suddenly reverse direction and swim velum first; others swim velum first directly off the bottom, sailing majestically surfaceward.

We confirm the observations of Turner and George (1955) that even when crowded to densities of fifty larvae per cubic centimeter, the larvae seldom collide with each other, for "the currents provided by the swimming organ seem to act like the slip stream of an aeroplane." In more dense cultures collisions are frequent, and some of the colliders fall toward the bottom. Those hitting the bottom frequently turn rapidly in place before swimming away.

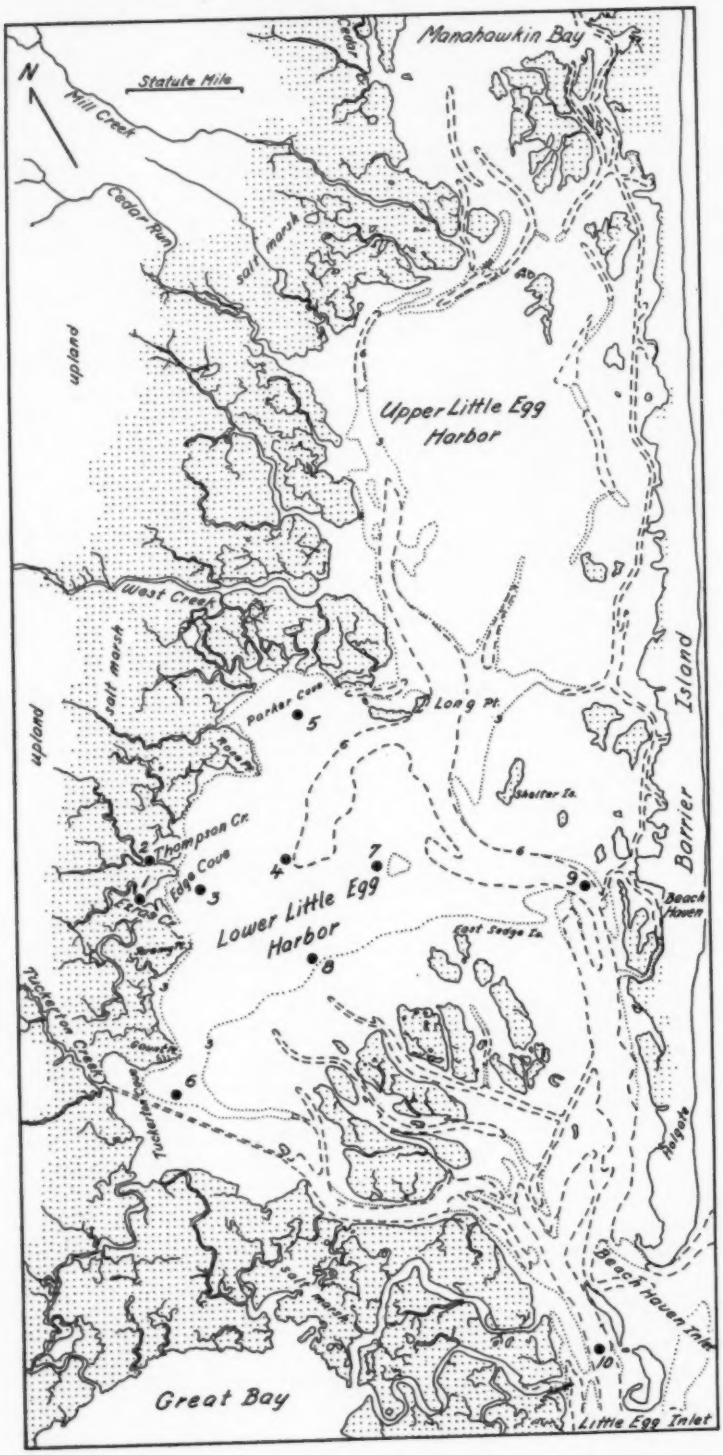
Mild pipette-driven currents of water and gentle knocks on the walls of culture containers do not bring about cessation of swimming, whereas a drop of water falling on the surface of a culture dish will stimulate larvae in the close vicinity of the splash to close their valves and sink for a short distance before they resume swimming. When they are swirled into a heap in the center of a small concave vessel, swimming veligers continue moving and soon disperse more or less uniformly throughout the water. So far as could be determined, healthy well-fed larvae never rest on the bottom of the culture containers; apparently they pass the entire veliger stage suspended in the water. Conversely, underfed larvae, or "diseased" larvae, drop to the bottom after a few days and remain there with only sporadic swimming efforts. In rearing larvae in still water in shallow open trays, we have observed that those (particularly the early stages) near the glass-water-air interface become trapped in the drying film there and may be killed by desiccation and increasing salinities.

Our fastest-growing veligers metamorphosed and started setting eight days after fertilization of the ova.

Planktonic Stages in the Field

Behavior and ecology of veligers of *M. mercenaria* were studied in two separate ecologically distinct estuaries. The first—Little Egg Harbor, New Jersey—is a large estuary characterized by dense populations of adult *M. mercenaria*, high uniform salinities, and moderate exchange and negligible flushing rates which contributed to retention of a high percentage of larvae to the setting stage. The second—Home Pond, New York—is a small intertidal arm of Gardiners Bay, and is also characterized by dense populations of *M. mercenaria*, high uniform salinities, and negligible flushing rate, but possesses exchange ratios as high as 73 per cent, so that young larvae originating within the pond are quickly transported out of it, and larvae which set in it enter on the late flood tide from the bay as pediveligers. The Home Pond studies have been published (Carriker, 1959); a report of those in Little Egg Harbor constitutes this section of this paper.

The first phase of this investigation involved study of a number of the more obvious environmental factors which might be associated with the behavior and ecology of early stages of *M. mercenaria*. Of these, morphometry, rainfall, tidal currents, water movements, salinity, and temperature were studied in detail; and wind direction and velocity, turbidity, and sediments were observed more generally. L. R. Pomeroy and H. O. Werntz aided materially in the collection of these hydrographic data.


Manahawkin Bay
N
Statute Mile
Mill Creek
Cedar Run
salt marsh
upland
Upper Little Egg Harbor
West Creek
salt marsh
upland
Parker Cove
Long Pt.
Shelter Is.
Barrier Island
Thompson Cr.
Edge Cove
Lower Little Egg Harbor
East Sedge Is.
Beach Haven
Tuckerton Creek
Great Bay
Little Egg Inlet
Beach Haven Inlet
Ridgeway
1
2
3
4
5
6
7
8
9
10

1. Hydrography of Little Egg Harbor

MORPHOMETRY, RAINFALL, AND EVAPORATION. Little Egg Harbor is a shallow coastal plain estuary of Type C (Pritchard, 1955) (Fig. 14) approximately four statute miles wide at the southwestern end and some ten miles long and possesses a water-surface area of 31.5 square miles. Upper Little Egg Harbor, with the exception of a few narrow, relatively deep channels, has a depth of one to two feet at mean low water and an area of 13.8 square miles. It extends northeastward beyond its boundary as Manahawkin Bay and beyond that as Barnegat Bay (Fig. 1). Little Egg Harbor is separated from the Atlantic Ocean by a continuous barrier island devoid of inlets except for the Beach Haven and Little Egg inlets at the southern extremity and Barnegat Inlet some eight miles northeast of Upper Little Egg Harbor. Tidal currents entering through Beach Haven-Little Egg inlets meet in the Manahawkin Bay area.

Lower Little Egg Harbor has an area of 17.7 square miles, and the deeper region (bounded by Long Point, East Sedge, and Gaunt Point) is characterized by average depths at mean low water ranging from four to seven feet. A cluster of salt-marsh islands hugs the southeastern corner of the bay; around the east of these runs a wide, deep channel, and lesser channels break through the islands and to the west of them. South of the islands, Little Egg Harbor communicates with the Atlantic by a channel about one mile in width which forks into the recently opened (1920) and relatively shallow Beach Haven Inlet, and farther south into the main and deeper Little Egg Inlet shared with Great Bay (Fig. 1). No sills are present. Maximum depth for the main channels is charted as 34 feet, but constantly shifting sands alter topography from year to year. Several small channels pass between Little Egg Harbor and Great Bay to the southwest.

Volumes of Little Egg Harbor (area delimited in Fig. 14) were determined by planimetry from U. S. Coast and Geodetic Survey Charts #825 and #826, from soundings on these charts, and from data (Table VII) on mean intertidal ranges in various parts of the bay:

Subtidal volume	4.09×10^9 ft.^{3}
Mean high-tide volume	5.88×10^9 ft.^{3}
Mean summer-spring high-tide volume (inclusive of the volume of water rising over the salt marsh above the mean high-water line, 0.25×10^9 ft^3)	6.49×10^9 ft.^{3}
Maximum summer-spring high-tide volume experienced during the summer of 1948, likewise including the volume of sea water over the salt marsh, 0.79×10^9 ft.^{3}	7.70×10^9 ft.^{3}

A marginal strip of salt marsh 0.5 to 3 miles in width, with a total area of 22.6 square miles, bounds Little Egg Harbor on the landward side. The barrier island to the southeast, ranging in width from 0.2 to 0.7 mile and composed mostly of sand, is now for the most part occupied by small towns, and there are also small towns along most of the streams in the Little Egg Harbor watershed. Despite this population, however, during the course of these studies the waters of Little Egg Harbor were free of domestic and industrial pollution.

The watershed draining into Little Egg Harbor, calculated planimetrically from New Jersey Division of Geology and Topography Atlas Sheets 32 and 33, has a total area of 77.2 square miles. This is 2.4 times larger than the water-surface area of Little Egg Harbor, and 1.4 times larger than the water-surface and salt-march areas of Little Egg Harbor combined. The watershed consists of relatively flat, sandy terrain clothed by typical pine-barrens vegetation (Buell and Cantlon, 1950) and drains into Little Egg Harbor through four streams of roughly similar size—Tuckerton Creek, West Creek, Cedar Run, and Mill Creek (Fig. 14). Salt marshes around the bay are closely dissected by shallow, meandering tidal creeks which usually terminate near the upland-salt-marsh boundary and transport little or no fresh water except during periods of heavy rainfall. Ezras Creek (used as a mooring for the "Cynthia" in 1948) and Thompson Creek (similarly used in 1949–51) are representative.

Since no data were available on the rate of discharge of fresh water into Little Egg Harbor, approximate values for runoff coefficients were obtained from available data (U. S. Weather Bureau, 1948–51; U. S. Geological Survey, 1948–51) on precipitation (Toms River Station for the Cedar Creek watershed) and rate of discharge of fresh water from the Cedar Creek drainage area, which measures 56.0 square miles,

FIG. 14. Bathymetric chart of Little Egg Harbor, New Jersey. Three- and six-foot contour lines are shown. Beyond the six-foot contour lines, depths range to a maximum of 34 feet in the main channel. (Chart traced from U. S. Coast & Geodetic Survey Charts Nos. 825 and 826.)

Table V

Computation of total volume of fresh water entering Little Egg Harbor per month during the summers 1948–1951.

Year and month	(A) Run off coefficient (cfs/sq. mi. watershed/ in. rainfall over Cedar Creek watershed).	(B) Average rainfall over Little Egg Harbor and watershed, inches	(C) Run off from Little Egg Harbor watershed into Little Egg Harbor, cfs.	(D) Evaporation from Little Egg Harbor and salt marshes, inches	(E) Average rainfall minus evaporation over Little Egg Harbor and salt marshes, inches	(F) Net volume of fresh water added to surface of Little Egg Harbor and salt marshes, cfs.	(G) Net volume of fresh water added to Little Egg Harbor from run off and rainfall less evaporation, cfs.
1948 June...	0.74	5.94	339.3	3.7	0.2	107.6	446.9
July...	0.52	3.49	140.1	5.4	−1.9	− 88.2	51.9
Aug....	0.35	9.49	256.4	4.3	5.2	242.6	499.0
1949 June...	7.30	0.28	157.8	6.0	−5.8	−278.2	−120.4
July...	0.63	2.27	110.6	6.5	−4.2	−197.5	− 86.9
Aug....	0.13	3.91	38.0	4.5	−0.6	− 27.2	10.8
1950 June...	0.51	2.47	97.2	5.3	−2.8	−135.7	− 38.5
July...	0.36	2.43	67.5	5.0	−2.8	−120.6	− 53.1
Aug....	0.24	2.64	48.9	5.1	−2.5	−117.3	− 68.4
1951 June...	0.47	4.83	175.2	5.4	−0.6	− 28.6	146.6
July...	0.40	2.62	80.9	5.8	−3.2	−150.6	− 69.7
Aug....	0.35	1.36	36.7	5.7	−4.3	−202.2	−165.5

at the Lanoka Harbor gauging station. Calculations were patterned after similar ones made by Pritchard and Bunce (1959). The Cedar Creek watershed is quite similar to that of Little Egg Harbor, and the two are only four miles distant at their closest boundaries. Based on these data, the Cedar Creek runoff coefficients provided the rate of discharge of fresh water in cubic feet per second, per square mile of drainage area, per inch of rainfall, for each of the three summer months of the period 1948–1951. On the basis of the known area of the Little Egg Harbor watershed, of observed mean precipitation over and near the watershed during the summer months, and of the runoff coefficients for Cedar Creek, we could compute the fresh-water runoff into Little Egg Harbor (Column C, Table V). Precipitation data were averaged from available U. S. Weather Bureau rainfall data at the Tuckerton, Bass River State Forest, Pleasantville, and Atlantic City stations and from precipitation data collected near the "Cynthia" starting in August, 1949 (Fig. 15: 1948–1951). In addition, from runoff data and precipitation directly on the surface of the water and salt marshes of Little Egg Harbor, and from loss of fresh water from Little Egg Harbor and its salt marshes by evaporation (based on U. S. Weather Bureau evaporation data for Pleasantville), we estimated the net volume of fresh water moving into, or out of (by evaporation), Little Egg Harbor for each of the summer months of 1948–1951 (Table V).

As Column G of Table V indicates, the net addition of fresh water to Little Egg Harbor from runoff and precipitation less evaporation varied considerably from month to month. Maximum movement of fresh water into the bay—499 cubic feet per second—took place in August, 1948 (Fig. 15: 1948). This contribution was roughly equivalent to 2.23×10^8 cubic feet (or 0.04 per cent of the mean high-tide volume of Little Egg Harbor) per tidal cycle of 12.4 hours; or 3.01×10^7 (0.52 per cent) per week, the minimum duration of the planktonic stage of clam larvae observed in Little Egg Harbor. At this rate of flow, it would take some 45 months for fresh water to replace sea water in the mean high-tide volume of the bay.

Conversely, during drier months, evaporation exceeded inflow of fresh water, bringing about a net flow of sea water into the bay. During the period of minimum precipitation—August, 1951 (Fig. 15: 1951)—a net total of 165 cubic feet per second of fresh water was lost from the bay and adjacent salt marshes by evaporation. This was equivalent to a loss of 7.40×10^8 cubic feet (or 0.01 per cent of the mean

high-tide volume of Little Egg Harbor) per tidal cycle, or 9.99×10^9 cubic feet (0.17 per cent) per week. At this rate of net loss of fresh water, the mean high-tide volume of Little Egg Harbor would be removed by evaporation in about 150 months.

TIDES. Tide level was recorded during each of the four summers on a portable automatic tide gauge (Coast and Geodetic Survey, 1941) secured to a platform supported on braced piling stuck deep in the bottom. In Ezras Creek (Station 1, Fig. 14) in 1948 the gauge was placed in the middle of the creek seventy-five feet inside the mouth, and during the remaining three summers near the middle of Thompson Creek (Station 2, Fig. 14) 135 feet inside the mouth of the creek. In both locations protection of the gauge from wave action from the open bay permitted the production of smooth curves on the tide gauge records. It was not feasible to refer our observed tide-level recordings to a tidal bench mark; therefore each summer the recording stylus of the gauge was set at an arbitrary level which would accommodate the range of local tides but not necessarily correspond to observed tide levels of other summers. Tide-height data were used principally to relate distribution of larvae of *M. mercenaria* to tidal phenomena.

The type of tide observed in the Edge Cove area was uniformly semidiurnal, the principal variations following changes in distance and phase of the moon. The height of high water was maximal after new moon and considerably lower after full moon. Curves representing high- and low-water heights paralleled each other closely (Fig. 15: 1948–1951).

In order to facilitate comparison, even though crude, of tidal amplitudes during the four summers, we averaged observed low-water heights (read from tide records to nearest 0.05 foot) for each summer to provide mean low-water levels; these levels were considered "zero height" and other tide-level figures were equated to these common planes. The mean high water for each summer was obtained by averaging high-water levels; maximum (highest) and minimum (lowest) levels for each summer represent single observations, all similarly equated to zero mean low water. These data are summarized in Table VI. Mean amplitude of the tides over the four-summer period was 1.85 feet; and maximum amplitude, in 1948, was 3.75 feet. In the Edge Cove area, the water level rose over the edge of the salt marsh at a tide-gauge height (equated to mean low water at zero) of 2.4 feet during the summer of 1950 and 2.3 feet during the summer of 1951 (Fig. 15: 1950, 1951).

The figures in Table VI for maximum, minimum, and mean high water show a consistent decrease in height from 1948 through 1951. Total rainfall in the Little Egg Harbor area during the periods in which tidal records were collected (Table V) was as follows: 1948, 18.92 inches; 1949, 6.46 inches; 1950, 7.54 inches; 1951, 8.81 inches. This trend in amplitudes may possibly be explained in part by variation in rainfall.

Decreasing tidal amplitude from the head of Little Egg Harbor toward the inlets is given in Table VII (compare Fig. 14). All figures, except those for Edge Cove, were taken from *Tide Tables East Coast North and South America*, U. S. Coast and Geodetic Survey.

WATER MOVEMENTS. Movements of water masses in Lower Little Egg Harbor were followed principally by means of a light current cross consisting of four vanes each one-square foot in area. In operation the cross was sus-

Table VI

Tide levels, in feet, for the Edge Cove area, Little Egg Harbor, N. J., for the summers of 1948–1951. Figures for each summer are equated to a mean low-water height for that summer of zero.

Tide Level	1948 June 23–Sept. 4	1949 June 5–Aug. 31	1950 June 20–Sept. 4	1951 June 15–Aug. 28	Four-summer mean
Maximum High...	3.21	2.82	2.70	2.77	2.87
Minimum High...	0.91	0.77	0.75	0.57	0.75
Mean High.....	1.96	1.87	1.80	1.79	1.85
Mean Low.....	0	0	0	0	0
Minimum Low...	0.61	0.92	0.85	0.52	0.72
Maximum Low...	−0.54	−0.68	−0.60	−0.83	−0.66

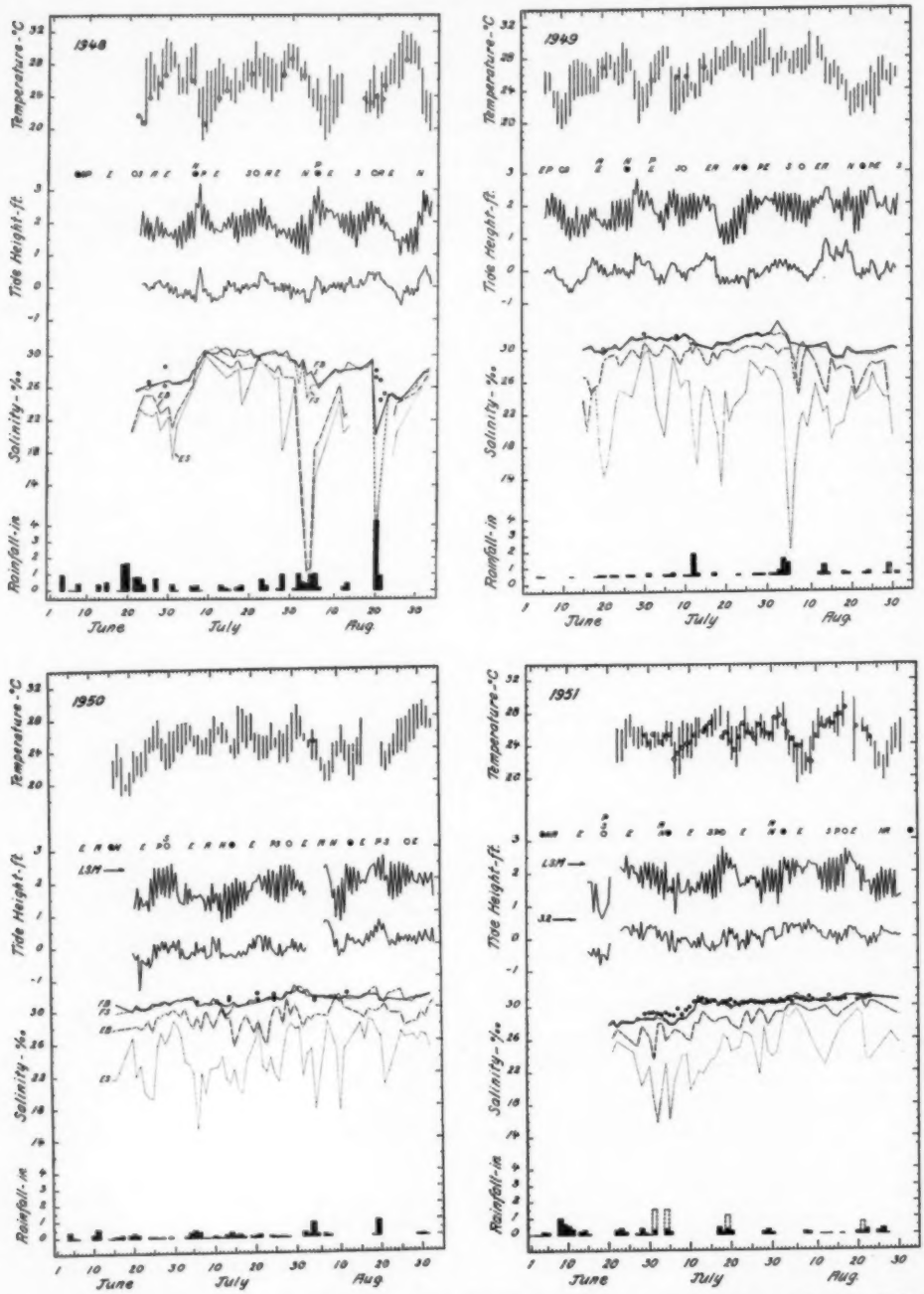


FIG. 15. Mean rainfall over Little Egg Harbor and its watershed, and tidal amplitude, salinity, and temperature of water in Ezras Creek (summer of 1948) and in Thompson Creek (summers of 1949–1951). Tide heights are equated to the observed mean low water level for the four summers of 1948–

Table VII

Mean and spring range of the tide at various points in Little Egg Harbor.

Area	Range, ft.	
	Mean	Spring
Manahawkin Bay	1.5	1.8
Edge Cove	1.8	2.1
Long Point	2.2	2.7
Beach Haven	2.2	2.7
Tuckerton Creek entrance	2.4	2.9
Holgate	2.6	3.1
Inlet (Tucker Island)	3.7	4.5

pended, the top about three feet below the surface of the water, from a small float; thus it hung about mid depth in much of the central basin of Lower Little Egg Harbor where maximal concentrations of clam larvae were encountered. The current cross was launched near low or high water in the early morning in order to obviate the effect of frequent strong afternoon winds and then followed in the laboratory powerboat as it floated from one slack period to the next. Hourly positions along the course of each run were plotted from bearings taken on fixed, charted landmarks with the aid of a magnetic compass and azimuth circle mounted in the boat. Approximately at each hourly fix, two or more vertical current velocity, temperature, and salinity observations were also made. In two of the seven tidal excursions which were followed (9–1–50 and 8–29–50, crossed circles, Fig. 16), the movement of surface water marked with fluorescein was studied simultaneously with that of deeper water by the current cross. The course of these seven runs is illustrated in Figure 16 and accompanying tidal, salinity, temperature, and current velocity data are tabulated in Table VIII.

Current velocities and directions were measured with a calibrated Ekman current meter at various depths throughout Lower Little Egg Harbor during the four summers. A composite of current patterns and maximum velocities observed during mid through late flood are presented schematically in Figure 17a, and current patterns for approximately mid ebb tide in Figure 17b. Current velocities exceeding those given in Figure 17 may have occurred, since it was not possible to measure velocities during maximal flow of the highest spring tides.

FIG. 16. Movement of water masses in Little Egg Harbor. Circles along the path of movement indicate hourly intervals, as labeled; arrows pointing up-bay indicate flooding tide; arrows pointing down-bay, ebbing tide; open circles, use of a cross suspended approximately 4 ft. below the surface of the water; crossed circles, use of fluorescein at the surface. Each tidal run is identified by the date on which the observations were made.

During early flood, sea water moving up the main channels from Beach Haven-Little Egg inlets was divided by the cluster of marsh

1951 (Table VI). Positions of the moon: ●, new moon; O, full moon; E, moon on equator; N or S, moon farthest north or south of the equator; A or P, moon in apogee or perigee. Salinity and temperature of the water at Station 4 superimposed over corresponding curves for salinity and temperature of water in creek (principally in 1951). Level of salt marsh at Edge Cove (LSM) is related to tidal levels (1950, 1951). Broken-line histograms over solid histograms, summer of 1951, refer to rainfall measured in rain gauge at the "Cynthia" for the Thompson Creek area when rainfall there exceeded the mean rainfall for the Little Egg Harbor watershed, indicated by the solid histograms.

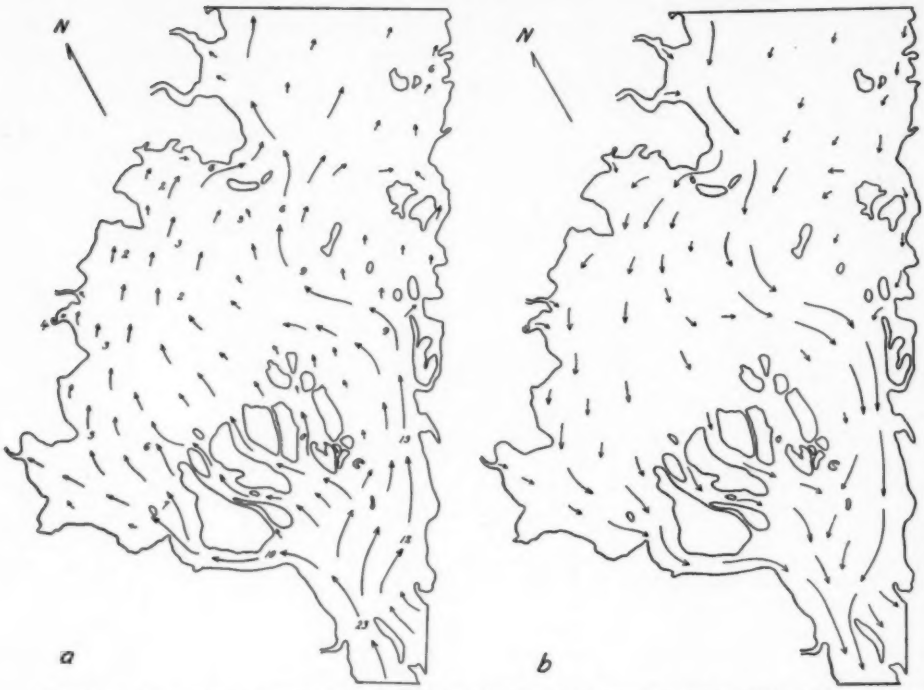


FIG. 17. a. Direction and maximum observed velocities of tidal currents in Lower Little Egg Harbor during mid through late flood tide. Length of arrows is roughly proportionate to current velocity, and numbers indicate maximum observed current velocities in ft./min. (to nearest ft.) between surface and mid depth. b. Direction of currents in Lower Little Egg Harbor during mid ebb.

islands which partially block the southern end of the bay: a small quantity flowed northwestward toward Tuckerton Creek and thence northeastward along the western shore of the bay toward Long Point; the greater volume moved northeastward along the barrier island about three miles and then swept windingly in a northward direction across the bay toward Long Point (Fig. 17a, 14). As the level of the flooding tide continued to rise, water found new passageways over shallower channels and shoals along the southern limits of the bay among the marsh islands and moved northward at the surface over deeper strata, flowing toward the center of the estuary from the southeast and southwest. The morphometry of the bay thus creates a complex circulation pattern in the central basin of Lower Little Egg Harbor where direction and velocity of currents continue to change throughout the vertical column as the tide runs its cycle. On the ebb the flows were reversed (Fig. 17b).

During low neap summer tides (tide level 1.2 feet at the tide gauge) approximately 20.4 per cent, and during mean summer high-tide periods (1.8 feet at tide gauge) about 30.3 per cent, of the sea water in Little Egg Harbor at mean high water emptied into the Atlantic Ocean on the subsequent ebb tide. During mean summer-spring high tides (2.1 feet at tide gauge) 36.8 per cent, and during maximum summer-spring high tides (3.2 feet at tide gauge) 46.8 per cent was exchanged.

SALINITY. The first phase of this study involved daily collection of water samples from the "Cynthia" in Ezras Creek (1948) and in Thompson Creek (1949–1951) at the surface and just off the bottom at low and high slack water; it provided salinity reference curves (Fig. 15: 1948–1951) to which salinities in Lower Little Egg Harbor were related. The second, likewise undertaken over the four-summer period, involved sampling water in Lower Little Egg Harbor (a) at several vertical levels at different stages of the tidal cycle at a number of stationary stations; (b) following and sampling moving masses of water at several

vertical levels from one slack period to the next; and (c) sampling horizontal transects over various portions of the bay as rapidly as possible within short portions of a single tidal cycle. Salinity samples were also taken simultaneously with most plankton samples. At the "Cynthia" surface water samples were dipped directly from the creek. Bottom samples were taken in a glass bottle perforated at the base and fastened to the end of a long glass tube; the flow of sea water into and out of the bottle was controlled by opening and closing of the upper tube. This simple device (first described to me by T. C. Nelson) permitted rapid collection of water samples immediately over the bottom. Away from the "Cynthia" subsurface water samples were pumped through a hose suspended at desired levels. Salinity was determined in the laboratory by means of Harvey's modified silver nitrate titration, with an accuracy of ±0.05 °/oo.

Salinity of sea water collected in the creeks from the "Cynthia," plotted as separate curves for bottom and surface, at both high and low water, is correlated with rainfall and tidal height in Figure 15. Close approximation of curves for bottom and surface flood slack water indicates thorough mixing of flood water entering these creeks. Salinities of sea water collected concurrently at Station 4 in Little Egg Harbor (Fig. 18) at all stages of the tides from surface to bottom coincided closely with those of both flood slack surface and flood slack bottom at Stations 1 and 2 and permitted use of the flood slack salinity data of the creeks as a guide to the level of salinities in Lower Little Egg Harbor.

Heavy precipitation in June and August, 1948, and June, 1951 (Fig. 15) depressed salinities slightly throughout Little Egg Harbor and steeply in the creeks. Ezras Creek is a tidal stream receiving no drainage from the land and thus the degree of dilution of sea water in it was related to the quantity of rainfall immediately over it and surrounding salt marsh (Fig. 15: 1948). Thompson Creek, on the other hand, receives fresh water from a small watershed 0.9 square mile in area, and lowering of salinity in it resulted from both direct rainfall and stream flow (Fig. 15: 1949–1951). Occasionally rainfall from small thunderstorms over the Thompson Creek watershed depressed salinities in the creek but was so localized that it was not registered on any of the existing rain gauges of the area (see especially August 10, 1950, Fig. 15). Fresh water ebbing from these creeks soon mixed with sea water in Little Egg Harbor, as demonstrated by the much higher salinities and virtual lack of stratification in the creeks during the late flood tide. From time to time spring high tides in the creeks were accompanied by higher salinities than normal (see especially the record high tide of July 8, 1948, Fig. 15); if not for the diluting effect of precipitation, increases in salinity would be detectible during all spring high tides because of the movement of bay water farther into the salt marshes.

One of the striking hydrographic features of Little Egg Harbor was the combined vertical and horizontal uniformity of salinity in the summer. During the four-summer period of these studies the maximum range in the vicinity of Station 4, inclusive of all stages of the tide

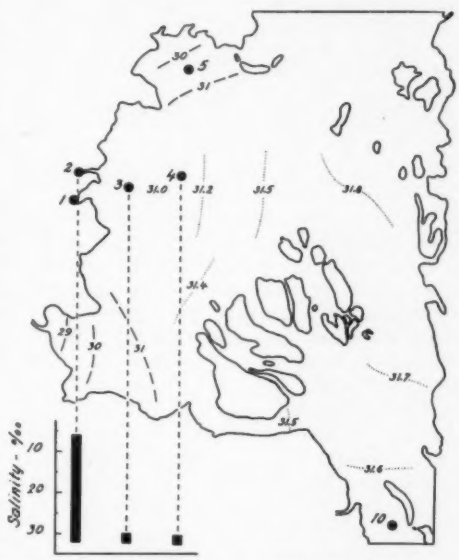


FIG. 18. Distribution of salinity in Little Egg Harbor. Isohalines represent average salinity in the vertical column of water at approximately mid tide when salinity in the vicinity of Station 4 was equivalent to 31.0 o/oo. Histograms at lower left depict range of salinity in the vertical column at ebb and flood slack over the four summers of 1948–1951 at Stations 2, 3, and 4.

and all vertical levels from surface to bottom, was only 8.8 °/oo (24.4 to 33.2 °/oo), while in Ezras and Thompson creeks it was 32 °/oo (see histograms, Fig. 18). This homogeneity may be explained by the fact that a proportionately small volume of fresh water enters the bay and is rapidly mixed by complex tidal currents moving over a broad, shallow basin and

by strong prevailing southwest winds. Figure 19 depicts vertical distribution of salinity from low to high water at three representative widely separated locations along the main axis of Lower Little Egg Harbor. At only one stratum, one-hour flood at Station 5, did the vertical range of salinity exceed 0.5 °/oo. Greatest vertical mixing took place at maximum flow of the current at mid flood and mid ebb. Slightly higher salinities entered Stations 10 and 4 from down-bay during the late flood, whereas at Station 5 salinities dropped slightly during the flooding tide as a mass of water from the Edge Cove area, slightly diluted by Thompson Creek water, moved toward Parker Cove (Fig. 14). Horizontal distribution of salinity in the bay is plotted in Figure 18. Isohalines represent average salinities in the vertical column at approximately mid-tide when salinity in the vicinity of Station 4 (the principal larval sampling station in the bay) was equivalent to 31.0 °/oo. This level of salinity, ± 1 °/oo, prevailed during July, 1948, most of the summer of 1949, all of the summer of 1950, and July and August, 1951 (Fig. 15). Slightly lower salinities occurred in the Parker Cove and Tuckerton Cove areas, depressed by fresh-water flow from West Creek and Tuckerton Creek respectively. A small, increasing gradient of salinity was present across Lower Little Egg Harbor, and a decreasing gradient, down the channel from Tuckerton Cove and down the eastern channel toward the inlet from Beach Haven. These isohalines indicate slight dilution of sea water along the southwestern shore of Lower Little Egg Harbor, the concentrating effect of evaporation in extensive shallow areas in Upper Little Egg Harbor, and very slight dilution by mixing with brackish water in Great Bay to the south in and off Little Egg Inlet. The last observation is corroborated by the fact that salinity did not rise appreciably in Little Egg Harbor during dry summer months when evaporation exceeded inflow of fresh water from the Little Egg Harbor watershed (Table V), fresh water being contributed by the Mullica River via Great Bay and mixing in the joint Little Egg Inlet. As precipitation increased over the Little Egg Harbor area, with consequent acceleration of flushing, salinities in the western portion of the bay dropped, and the gradient of salinity across the bay steepened. When the maximum net monthly rate of fresh water was added to Little Egg Harbor (August, 1948, Table V), contributing the equivalent of 0.04 per cent of the mean high-tide volume of the bay per tidal cycle, salinities at Station 3 dropped to about 25 °/oo. At the other extreme, during periods of minimal precipitation, fresh water was lost from Little Egg Harbor at the rate of 0.01 per cent of the mean high-tide volume of the bay per tidal cycle (August 1951, Table 5), elevating salinities at Station 3 to approximately 31 °/oo.

It was not possible to determine net nontidal movements up-bay on the bottom and down-bay on the surface resulting from the flow of fresh water out of the bay, but vertical homogeneity of salinity throughout most of the bay during periods of minimal to moderate summer precipitation suggest that negligible nontidal movements existed then.

TEMPERATURE. The investigation of thermal properties of sea water in Little Egg Harbor was patterned after the investigation of salinity. In Ezras and Thompson creeks temperature of surface and bottom water was taken almost daily at ebb and flood slack water with a reversing thermometer. In addition a thermograph was installed on the tide gauge platform near the "Cynthia," and the mercury bulb was placed about a foot off the bottom in water approximately 4.5 feet deep at mean low water. In Little Egg Harbor temperatures were taken with a reversing thermometer as salinity and larval samples were collected.

Thermograph records for bottom water in the creeks for the four summers revealed the occurrence of thermal maximum in the late afternoon on sunny days; the thermal minima was in the early morning. These daily extremes, plotted in Figure 15, show a maximum range over the four summers of from 18.6° to 32.0°C., whereas the range recorded in the bay at Station 4 was from 18.6° to 28.6°C. Highest water temperatures in the creeks occurred when late ebbing tides coincided with hot, sunny afternoons. Because of the considerable distance of creek water from the inlet, about six statute miles across shallow Lower Little Egg Harbor, its thermal characteristics were most strongly influenced by insolation, and only slightly, if at all, by cooler flood water from the ocean.

Daily surface and bottom water temperatures (not plotted) taken at ebb and flood slack off the "Cynthia" revealed a maximal vertical thermal difference between surface and bottom over the four summers of 4.0°C. at ebb and 2.5°C. at flood slack. However, such extremes were uncommon. On the whole, vertical thermal mixing of creek water was thorough. For example, in 1951 (the year representative of the four summers) in only 5 of the 109 sets of

Table VIII

Tidal, salinity, temperature, and current velocity data collected during study of movements of water masses in Lower Little Egg Harbor as plotted in Figure 16.

Date of run	Difference in tide level between low and high tide at tide guage, feet	Duration of observation, hours	Range in surface salinity during observations, and maximum difference, o/oo	Range in surface and bottom temperatures during observations, and maximum difference, °C.	Maximum current velocity observed during run, feet/sec.
8-27-51	low to high: 1.65	7.0	31.22 to 31.29: 0.07	21.8 to 22.7: 0.9	2.16
9- 1-50	low to mid flood: 1.80	3.6	—	24.6 to 25.0: 0.4	1.21
8-23-51	low to high: 1.95	6.3	31.09 to 31.33: 0.24	23.7 to 24.7: 1.0	0.92
7-14-50	high to low: 1.20	6.2	31.27 to 31.41: 0.14	25.6 to 26.1: 0.5	0.20
8-13-51	high to low: 1.40	6.7	30.40 to 30.80: 0.40	26.3 to 26.9: 0.6	0.33
8-22-51	low to high: 1.95	6.5	30.82 to 31.76: 0.06	25.4 to 25.6: 0.2	0.36
8-29-50	low to high: 1.80	6.5	—	25.8 to 27.4: 1.6	0.92

ebb and flood-slack surface and bottom temperature readings recorded during the summer were differences between surface and bottom 1.4°C. or greater. In 30 of these sets no measurable vertical difference could be detected; 7 of these occurred at ebb slack and 23 at flood slack. These data support those on salinity, which demonstrate that vertical mixing of water in the creeks was most thorough during the late flood tide.

Vertical thermal stratification in Lower Little Egg Harbar during the four summers was negligible at all stages of tide (Fig. 21), and temperature of separate water masses followed in the bay from mid morning to late afternoon did not vary more than 1.6°C. (Table VIII, Fig. 16). Mean daily water temperatures for Station 4, superimposed on daily maximum-minimum temperatures of creek water recorded by the thermograph (Fig. 15), occupy an approximate mean position on curves from creek-water temperatures. Thermograph recordings thus provided a useful guide to water temperatures in the central basin of Lower Little Egg Harbor.

In Lower Little Egg Harbor an increasing thermal gradient radiated from the inlet toward Edge Cove and Upper Little Egg Harbor. This gradient was most pronounced during late flood tides of periods of maximal water tem-

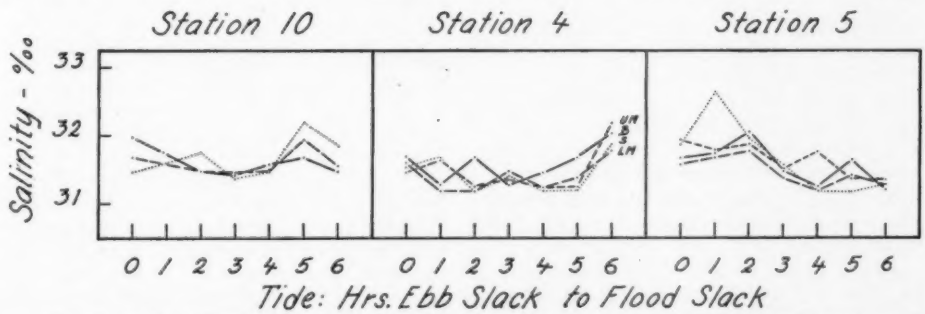


FIG. 19. Vertical distribution of salinity at Station 10 (inlet), August 8, in water 29.5 feet deep at low water; at Station 4, July 21, 8 feet deep; and at Station 5 (Parker Cove), July 25, 1950, 5 feet deep. S, surface; UM, upper middle stratum; LM, lower middle stratum; B, bottom salinity.

peratures in the Edge Cove area (Fig. 20). As the tide flooded, cool inlet water moved principally up the main channel of the bay, pushing the warmer water before it. On the ebb the one-degree isotherms spread as they moved down the bay toward the inlet. As

FIG. 20. Water temperatures, °C, in Little Egg Harbor during late flood tide on July 4, 1949, when tide level at flood slack registered 1.85 feet on the tide gauge. Maximum high tide that summer registered 2.82 feet (Table VI). Isotherms represent mean vertical temperatures.

insolation heat decreased, temperatures at the head of the bay and in the inlet approached each other. Data on horizontal distribution of water temperature complement those on movement of water masses in the bay (Figs. 16, 17) and demonstrate that for the most part cool inlet water flowed unidirectionally up and down the main channels, mixing only peripherally with that in the central basin of Lower Little Egg Harbor.

The thoroughness of vertical thermal mixing is illustrated in Figure 21 (compare with Figs. 14, 20). Representative stations 10, 7, 4, and 5 along the long axis of the estuary were sampled vertically at bottom, middle, and surface at hourly intervals from ebb slack to flood slack. On warm, sunny days the entire vertical column of water warmed slightly during the course of the day, even during the flooding tide. This is especially striking at Station 7 (compare with Fig. 15, for June 29, 1948), where observations were made on a hot day preceded by several cool ones.

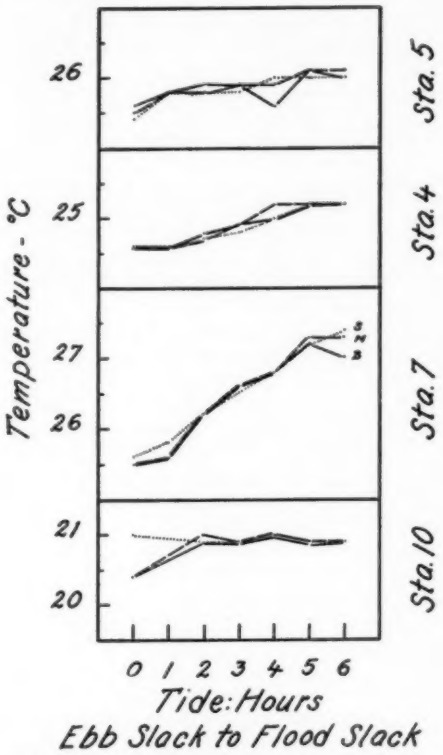


FIG. 21. Vertical distribution of temperature at Station 10 (inlet), Aug. 8, 1950, in water 29.5 feet deep at low water; at Station 7, June 29, 1948, 6 feet deep; at Station 4, July 21, 1950, 8 feet deep; and at Station 5 (Parker Cove), July 25, 1950, 5 feet deep, at approximately hourly intervals from ebb slack to flood slack. S, surface; M, middle; B, bottom temperature.

SEDIMENTS AND TRANSPARENCY OF WATER. Sediments in the central basin of Lower Little Egg Harbor consisted of firmly compacted fine sand and silt with a small percentage of organic detritus. Detrital content increased markedly in the direction of the salt marshes, bottoms becoming quite soft along the landward area of the bay. Sediments in channels and intertidal flats surrounding the marsh islands in the southern

part of the bay consisted of coarser clean sand. Bivalve shell fragments, mostly of *Crassostrea virginica* and *Mercenaria mercenaria*, were distributed irregularly over the bottom of the bay, most densely in areas along the northwestern section, where oysters were being cultivated. Except for scattered shell deposits, salt-marsh creek bottoms were composed of a firm central and soft marginal zones of fine, black, highly reducing organic mud.

Transparency was measured with a Secchi disk. During periods of calm to light wind, over the cycle of the tide, the disk disappeared from view at depths ranging from 0.6 to 1.2 meters (approximately 2 to 4 feet) at Stations 1 and 2 in the creeks, and from 1.2 to 1.8 meters (approximately 4 to 6 feet) at Station 4 in the central basin of Lower Little Egg Harbor. Transparency decreased as tidal flow increased in the tidal cycle. Bay waters, which received relatively little cedar swamp discharge, were noticeably free of stains. Generally clear water in the bay greatly facilitated collection, sorting, and study of plankton samples.

SUMMARY OF HYDROGRAPHY OF LITTLE EGG HARBOR. Float and current studies disclosed that in the central basin of Lower Little Egg Harbor the tidal excursion was relatively short as compared with that on the eastern side. Computations of net movement of fresh water into the bay during the summer indicated that insignificant flushing of bay water into the ocean fluctuated with the net flow of sea water into the bay during periods when evaporation rate exceeded the rate of fresh-water inflow. The movement of relatively small volumes of fresh water into the bay from all sources combined with a complex circulation pattern and wind action, produced relatively uniform high summer salinities throughout the bay and almost eliminated vertical salinity and thermal stratification. The uniformity of salinity suggests rapid and complete mixing of the water. Calculation of the volume of water in the bay at various stages of the tide showed that exchange was substantial. Thus the central basin of Lower Little Egg Harbor functioned somewhat as a retaining vessel of relatively homogeneous sea water in which a large proportion of clam larvae in many instances were held for at least the duration of the veliger stage.

2. Behavior and Autecology of Veligers in Little Egg Harbor

Our field study of veligers of *M. mercenaria* was designed to provide information on natality rate; quantitative seasonal, horizontal, and vertical distribution; and density, duration, movements, and mortality rate of the pelagic stages of the larvae in relation to several environmental variables: horizontal movements of water masses, current velocity and direction in various vertical strata, exchange, flushing, stage of the tide, horizontal and vertical distribution of temperature and salinity, light intensity and time of day, relative velocity and direction of the wind, general nature of the bottom, and density and distribution of parent *M. mercenaria* in the bottom.

DISTRIBUTION AND ABUNDANCE OF ADULT CLAMS. Before 1920 Little Egg Harbor communicated with the Atlantic Ocean by way of Little Egg Inlet through a narrow channel. Salinity in Little Egg Harbor was low, and bottoms were dominated by the oyster *Crassostrea virginica*. On February 5–6, 1920, a heavy storm cut a large, shallow additional inlet, Beach Haven Inlet, just north of the old one, thereby permanently accelerating exchange and elevation of salinity in the bay. Oyster production began a steady decline correlated with higher salinities and increasing quantities of sand which accumulated over the bottom (T. C. Nelson, personal communication). After this invasion of saltier water *M. mercenaria* became the dominant bottom organism in the sound and thereafter provided a livelihood for a considerable proportion of the local human population of Tuckerton and vicinity.

During the summers of 1948–1951 *M. mercenaria* were found in varying though dense concentrations in all bottoms of Lower Little Egg Harbor. Densest populations occurred in the open lower portion of the central basin of the bay immediately northwest of the marsh islands above the inlet (Fig. 14). The largest clams were found in tidal creeks and salt marsh ponds, though present in significantly less numbers. In the bay, clams were generally smaller in size and the condition of the flesh was not so good as that of those in the creeks. Continuous commercial tonging (power dredging was prohibited by state law) in the bay year after year probably reduced the maximum size of clams there. No explanation is available for the inferior meat of clams in the open bay, though Pomeroy (personal communication) has suggested an insufficient food supply to support such large populations. The relation of primary production to production of clams in this bay is worth studying, particularly in view

of the interesting hydrography and large populations of clams sustained there.

Data are available which show that Lower Little Egg Harbor has supported a high annual yield of *M. mercenaria* since at least 1929 (Fig. 22). Mr. E. Walter Parsons, Jr., a planter and shipper of oysters and clams in the Tuckerton area, has kept an accurate record of marketable clams passing through his shipping plant from Lower Little Egg Harbor and has permitted us to publish these data. He estimates that during the period 1929–1951 his plant received about one-tenth of the clams harvested commercially in this bay; thus his figures provide a rough index of the total annual production of clams there. The minimum harvest received by Mr. Parsons was 4,565,000 clams in 1932 and the maximum was 16,726,000 clams in 1946. The minimal total yield of the bay (yield to all clammers based on Mr. Parson's estimate) in 1932 represents an average take of 0.8 clam per square yard of bay bottom, while the average take for the maximum yield in 1946 was 3 clams per square yard per year. Whether trends in the curve of clam production over this twenty-two-year period (Fig. 22) represent fluctuations in tonging effort, in clam populations, or both is not known.

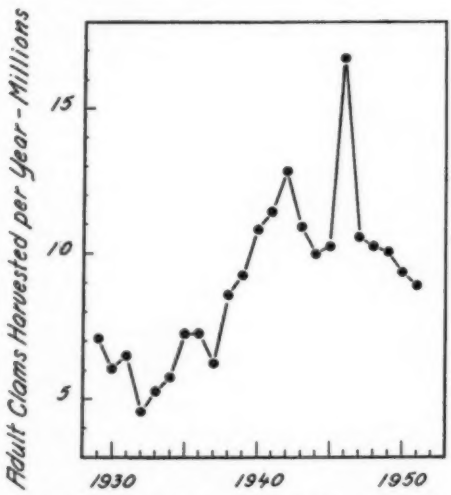


FIG. 22. Number of marketable *M. mercenaria* received per year by E. Walter Parsons' Shipping House, Tuckerton, New Jersey, from Lower Little Egg Harbor.

Abundant filter-feeding mollusks competing for food with *M. mercenaria* in Little Egg Harbor were: *Area campechiensis, Crassostrea virginica, Anomia simplex, Pitar morrhuana, Modiolus demissus, Ensis directus, Mya arenaria, Tellina tenera, Gemma gemma, Crepidula fornicata,* and in the lower reaches of the bay *Spisula solidissima* and *Mytilus edulis.* The following principal predators of post-setting stages of *M. mercenaria* were found: *Callinectes sapidus,* Xanthidae, *Polinices duplicatus, P. heros, Urosalpinx cinerea, Eupleura caudata, Busycon canaliculatum,* and *B. carica.* Moon snails and knobbed conchs were more abundant in the lower extremities of the bay, channeled conchs in the upper regions; crabs and oyster drills were distributed more or less throughout the bay.

It is worth emphasizing that clam production in Lower Little Egg Harbor, in spite of inroads by man and nonhuman predators, remained at a high level over a long period of time, producing the dense swarms of larvae which made possible these autecological studies.

METHODS OF COLLECTING AND STUDYING CLAM LARVAE. Plankton samples were pumped manually into a metal drum in the boat through a graduated intake hose five meters long by means of a large calibrated brass oscillating clock pump. The drum was outfitted with automatic siphons for draining. Sea water was poured into the drum through a standard conically shaped silk-bolting-cloth net suspended in sea water in the drum and equipped with a detachable screened bucket which permitted ready removal of plankton samples to bottles. The bulk of sampling was performed with 18 XXX silk-bolting cloth which retained the smallest shelled stages of clam larvae. For sampling older stages only 12 XXX silk-bolting-cloth nets were used. The intake of the hose was screwed into an opening in the center of a metal disk about six inches in diameter, opposed by a similar solid disk parallel to and held about an inch and a half from the first by a strip of one-eighth-inch mesh bronze screen soldered to the circumference of the two disks (Fig. 23). This basket excluded larger organisms from plankton samples and, more importantly, guided collection of larval samples from a relatively narrow vertical stratum of sea water. Lead weights secured to the intake basket steadied the hose in a vertical position in swiftly flowing water. A galvanized iron disk approximately eighteen inches in diameter with an upturned edge of about one-half inch was tied, when needed, under the intake basket of the hose to permit pumping plankton samples immediately off the bottom from an anchored

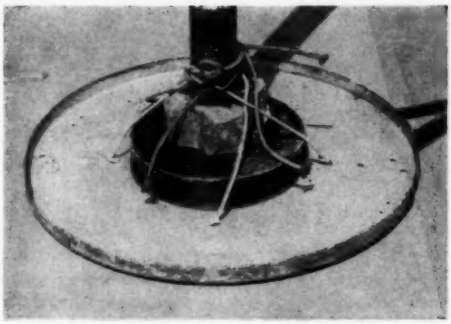


FIG. 23. Screened basket and bottom disk on intake of hose used in pumping plankton samples in Little Egg Harbor.

boat (Fig. 23). Samples were pumped after sediment thrown into suspension by the lowering of the gear had been removed by tidal currents. Single vertical samples were generally pumped while the boat floated over a station with the tide, whereas series of vertical samples were taken from the boat at anchor. The volume of plankton samples ranged from 25 liters when larvae were numerous to 1000 liters when ready-to-set larvae were scarce.

Routine sampling of larvae was carried out off the "Cynthia" at Stations 1 and 2 and in the bay at Stations 3 and 4 to provide a general picture of daily larval abundance. Horizontal distribution of larvae, obtained from transects over different portions of the bay, demonstrated that maximum concentrations of clam larvae occurred principally in the general area between Stations 4 and 8. This information, and the proximity of the point to the "Cynthia," provided the basis for location of our major sampling station at Station 4 (Fig. 14).

Series of vertical plankton samples from surface to bottom, in that order, during the cycle of the tide were taken mainly at Stations 4, 2, and 1. Each vertical plankton sample of the most complete vertical series was accompanied by observations on time of day, depth, current velocity and direction of current, temperature, and salinity; and the series as a whole was accompanied by stage of the tide, relative intensity of light striking the surface of the water, relative velocity and direction of the wind and thus degree of disturbance of the surface of the water, transparency of the water, nature of the bottom, and proximity to spawning parents.

With the apparatus described, and with fifty-liter samples at a time, two operators required approximately five minutes to complete observations at each stratum. Thus in any mass of water sampled serially in this manner, the horizontal spatial and temporal interval between the surface and bottom set of observations would depend upon the depth of water, the number of vertical samples taken, and the velocity of the current. This slanting of the vertical series should be kept in mind in the interpretation of data taken from them. In less complete vertical series, some of the observations accompanying the plankton sample were omitted. Ideally, simultaneous observations for all factors should have been made at all vertical strata, but this was not possible at the time of the study. To my knowledge this has not yet been achieved in estuarine planktonological studies and should be strived for in the future.

Plankton samples were flushed from the plankton net into stoppered glass bottles with a bulb pipette and killed and preserved by addition of neutralized formalin (full-strength formalin saturated with sodium borate) to make 2 per cent formalin in the plankton sample. This solution kills larvae quickly with soft parts retracted within the valves and does not bleach or alter the larval shell for several days. In samples containing much organic detritus, however, the *p*H of the preservative soon drops, bringing about erosion of the valves. Such "dirty" samples were usually examined first. Fluids for preservation of bivalve larvae for long periods of time have been described (Carriker, 1950). In the best of these, bivalve larvae free of organic detritus have retained glossy, clear valves and some pigment incorporated in the shells for about eight years. This fluid has the following composition, with a *p*H of 8:

Commercial formalin	5 cc.
Cane sugar	50 g.
$Na_2B_4O_7$ 10 H_2O, sodium tetraborate	0.25 g.
Filtered sea water, to make	500 cc.

Cleaning, sorting, identification, and counting of larvae of *M. mercenaria* were performed in the "Cynthia" laboratory as soon after sampling as possible. In formalin-treated practice plankton samples were washed with fresh water through a series of three nested stainless steel screens three inches in diameter with successive maximum diagonal pore dimensions of 340 μ (screen No. 60), 150 μ (No. 150), and 65 μ (No. 325). Clam larvae first appeared in the water as shelled veligers averaging about 95 μ in length and disappeared

from the water at lengths ranging from 180 to 200 μ. Thus all clam larvae passed through screen No. 60 but larger plankters and gross materials were retained. The contents of this screen were examined microscopically directly on the screen. Most umboned clam larvae, approximately 140 μ and longer, were held on screen No. 150. This fraction of the plankton with accompanying detritus was rinsed into a deeply concave glass swirling dish (such as the bottom half of a macro Kjeldahl flask). Fractions relatively free of organic detritus were examined without washing. Larvae were sorted and counted in the dish under a magnification of 34X. If detritus (usually in the form of minute fecal pellets) was abundant, as in samples pumped near the bottom, mollusk larvae and sand were swirled gently to the center of the dish with a small stream of water from a bulb pipette; detritus remaining at the periphery of the dish after the water came to a standstill was drawn off gently with another bulb pipette. Occasionally, as a check on the possible loss of larvae in the cleaning process, washings were allowed to settle in a settling cone and sediment in this was re-examined for veligers. Material retained on the finest screen (No. 325) was rinsed through a funnel into a 50 ml. stoppered graduate and the volume of water therein brought to 50 ml. The graduate was then shaken thoroughly to disperse bivalve larvae uniformly, and duplicate 5 cc. aliquot portions, each taken immediately after shaking, were drawn quickly from the middle of the graduate and examined in a shallow, concave glass dish under a magnification of 34X. A dissecting needle with tip filed flat on opposing sides facilitated sorting and counting. A multiple laboratory counter accelerated recording of the counts. Larval densities were expressed as number of larvae per 100 liters of native sea water. Examination of size fractions of the plankton samples in this manner has the advantage of making possible maximum accuracy of sorting and counting of the older, least numerous stages of clam larvae.

In all, during the four summers of 1948–1951, some 1,100 plankton samples were collected and examined.

Accurate identification of all stages of clam larvae in plankton samples was the most difficult aspect of our study in Little Egg Harbor. The close, sometimes bewildering similarity of a few of the developmental stages of some unknown species of bivalves to comparable stages of *M. mercenaria* in the bay forced us during the summers of 1947 and 1948 to undertake a preliminary systematic survey of the different species of bivalve larvae in the area (Carriker, 1950). This was done on the premise that recognition of the majority of these (if not to actual species at least to type) would minimize confusion of different stages of clam larvae with those of other closely related species. To this end all the different available kinds of bivalve larvae in the plankton were catalogued: in ten species, identification was possible by reference to the scanty literature; in eleven other species in hand identification was not possible, and provisional names were applied. Representative stages of the different species were measured, sketched, and preserved, and main diagnostic features were recorded. On the basis of these preliminary data, we constructed an incomplete taxonomic key to the bivalve larval types in the bay. Reference to this key, to preserved bivalve larval types, and to clam larvae cultures in our laboratory early in the summer of 1948 from gametes of known parents made possible positive identification of the larvae *M. mercenaria* in the plankton after July 14, 1948.

TEMPORAL DISTRIBUTION OF CLAM LARVAE. Loosanoff (1954) has demonstrated that specimens of *M. mercenaria* in the laboratory are able to develop normal gonads and to spawn repeatedly at approximately six-month intervals independently of day length, tidal rhythm, precipitation, kinds of plankton, and other factors present in the late spring and early summer when gametogenesis occurs normally. Spawning is induced in the laboratory, at least, by thermal and chemical stimuli. Although the factors that stimulate spawning under natural conditions are still obscure, it appears that in Little Egg Harbor spawning was triggered at least in part by thermal stimulation.

Seasonal distribution of clam larvae, associated with spawning time of swarms, daily median water temperature, and daily mean high-tide level is given in Figures 24 through 27. High-tide level is based on a mean low-water level of zero, and will be referred to hereafter as tidal amplitude. Other associated data on precipitation, salinity, maximum-minimum water temperatures, and daily low- and high-tide levels are presented in Figure 15.

Each summer the largest, most dense swarms of clam larvae appeared during the month of July: July 28–29, 1948; July 5, 1949; July 5–6, 1950; July 9–12, 21–22, 1951. The number of spawnings varied considerably from

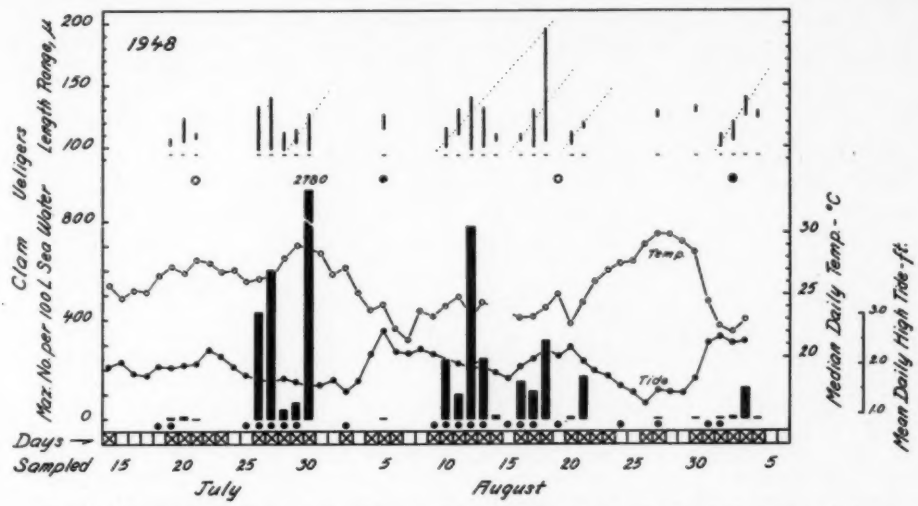


FIG. 24. Seasonal distribution, abundance, and range of length of veligers of *M. mercenaria* in upper Lower Little Egg Harbor (Stations 1–4) in the summer of 1948. Larval densities represent maximum number of larvae of all sizes per 100 liters of sea water at any of the vertical strata sampled per day; densities greater than 800 per 100 liter are expressed numerically at the top of the broken bar in the histogram. The date of spawning of each swarm of larvae (approximately one day prior to first appearance of earliest straight-hinged veligers) is indicated by solid circles under the histogram. Curves for median daily water temperatures (from thermograph recordings) and for height of mean daily high tide (from tide gauge recordings) are plotted near bottom of figure. See Figure 15: 1948, for additional environmental information. Length ranges (maximum and minimum lengths of larvae in a representative aliquot portion of each plankton sample) suggest, where there are sufficient data, the rate of growth (dotted diagonal lines) of individuals in each swarm based on the more complete data of the single isolated swarm of June 24–July 1, 1950 (Figs. 26, 31). The broken base line under length ranges represents length of shortest larvae found in plankton, 93 μ. Frequency of sampling is indicated by the crossed squares at the bottom of the figure.

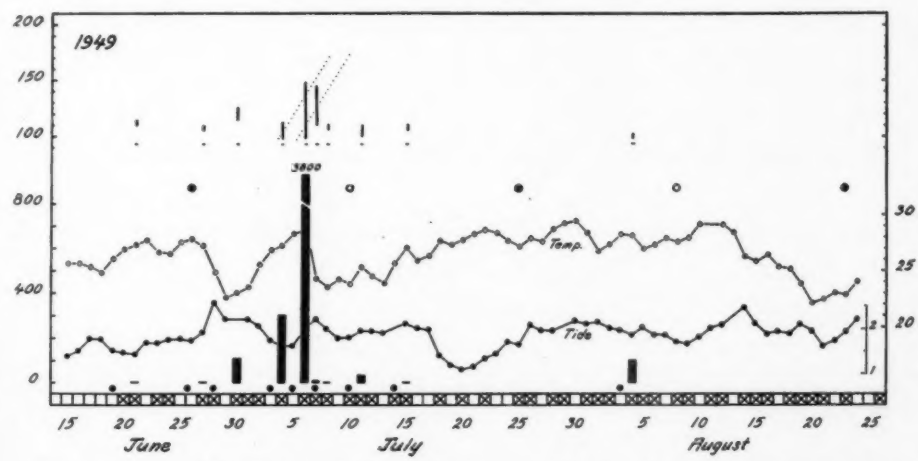


FIG. 25. Seasonal distribution, abundance, and range of length of veligers of *M. mercenaria* in upper Lower Little Egg Harbor (Stations 2–4) in the summer of 1949. Remainder of legend as in Figure 24. See Figure 15: 1949, for other environmental information.

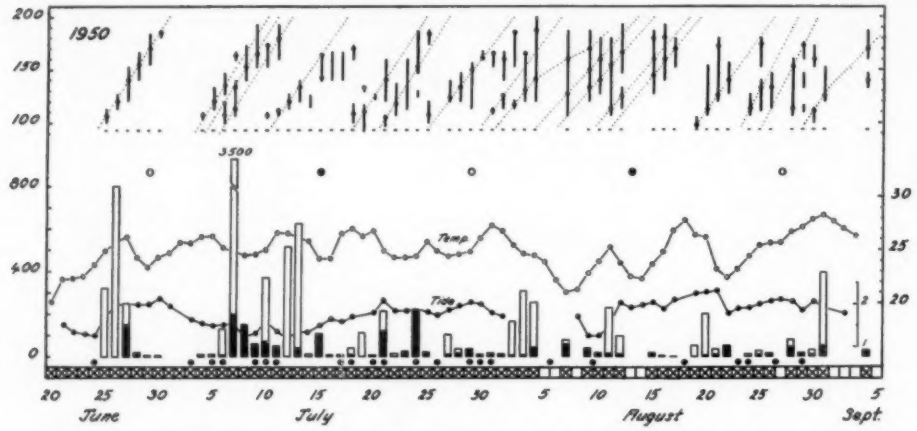


FIG. 26. Seasonal distribution, abundance, and range of length of veligers of *M. mercenaria* in upper Lower Little Egg Harbor (Stations 2–4) in the summer of 1950. Each open circle joined by dotted diagonal lines at the top of the figure represents the length of the most abundant size group(s) in the sample. Open histograms represent clam larvae ranging in length from 93 to 140 μ; solid histograms, represent 141 μ to setting stage. Remainder of legend as in Figure 24. See Figure 15: 1950, for other environmental information.

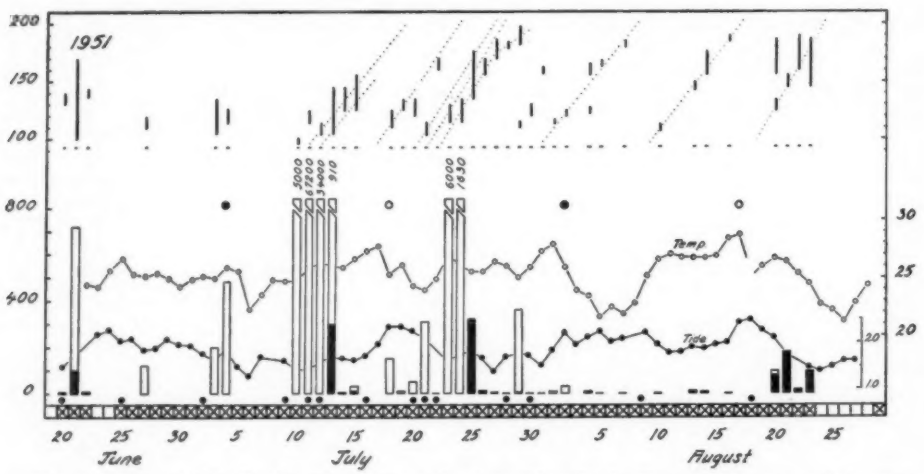


FIG. 27. Seasonal distribution, abundance, and range of length of veligers of *M. mercenaria* in upper Lower Little Egg Harbor (Stations 2–4) in the summer of 1951. Remainder of legend as in Figures 24 and 26. See Figure 15: 1951, for other environmental information.

summer to summer, decreasing in the following order: 1950, 1948, 1951, 1949. The fewest larvae were found during the summer of 1949, and maximum densities in 1951. Landers (1954) likewise found differences in the time of occurrence and duration of spawning of *M. mercenaria* in Narragansett Bay, Rhode Island. He suggests that variations in spawning patterns within and between seasons may be caused by variations in water temperature, early higher-than-average temperatures concentrating and advancing the spawning period and consistently low seasonal temperatures retarding and prolonging spawning at a high level. In a general way, Landers' hypothesis applies in part to the two summers in Little Egg Harbor in which minimal and maximal spawning was observed. The prolonged high-intensity spawning in the

summer of 1950 (Fig. 26) was accompanied by low temperatures in early June and moderate temperatures throughout the rest of the summer. By contrast, what little spawning was observed in 1949 (Fig. 25) was concentrated in the early part of the summer during a period of moderate water temperatures, and thereafter, when almost no spawning occurred, water temperatures remained high. Abundance and frequency of spawning is probably not explained on the basis of temperature alone, but temperature probably is one factor.

Of the six major spawnings which occurred in Little Egg Harbor during the four summers followed by maximum densities of over 1,000 larvae per 100 liters of sea water, five (July 29, 1948; July 5, 1949; July 5, 1950; and July 9 and 11, 1951) took place in a period of two or more days of rising median daily water temperatures; and one (July 21, 1951) coincided with falling median daily water temperatures (Figs. 24–27). Analysis of all spawnings over the four-summer period (Fig. 28) indicates that 73 per cent of a total of sixty-seven spawnings occurred on rising median daily water temperatures and twenty-seven per cent of falling temperatures. The average median daily temperature for spawnings which took place on rising temperatures was 26.2°C.; for spawnings on falling median daily temperatures, the average was 24.4°C., giving an over-all average median daily spawning temperature of 25.7°C. These observations support those in the laboratory, where we obtained both artificial and spontaneous spawnings on both rising and falling temperatures. Seventy-two per cent of the spawnings (Fig. 28) took place within six-degree days (sum of number of degrees of change per day) following a peak or trough in the curve for median daily water temperature, suggesting that only slight changes in temperature stimulate spawning after gonads are physiologically mature and the animal is ready to discharge gametes. The density of larval swarms bore no apparent relationship to the number of degree days between the time of spawning and the previous median daily thermal change (Fig. 28).

Median daily spawning temperatures encountered for swarms of all densities during the four summers were approximately the same, 25.7°C. (Fig. 29b). Spawnings occurred

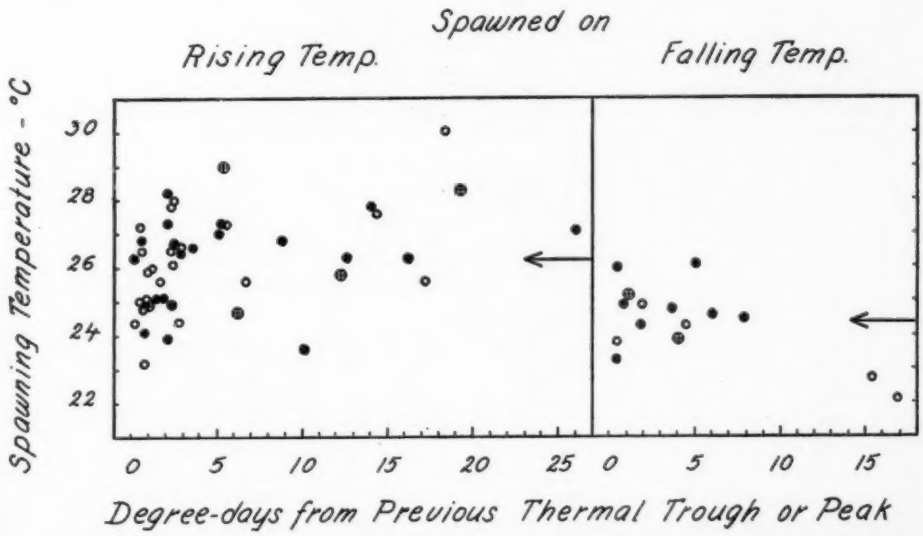


FIG. 28. Comparison of spawning of *M. mercenaria* in Little Egg Harbor on rising and on falling median daily water temperatures (compare with data in Figures 24–27) during summers of 1948–1951. Median daily water temperature at which spawning occurred is plotted against number of degree-days intervening between the previous trough in the thermal curve and the spawning temperature (for spawnings occurring on rising temperatures) and between the previous peak and spawning temperature (or spawnings occurring on falling temperatures). Maximum daily density of larval populations is expressed as follows: ○, 1–100; ●, 101–500; ⊙, 501–1000; and ⊕, 1001–67,000 early straight-hinged larvae per 100 liters. Degree-days refer to total number of degrees rise or fall per day in the curve for median daily temperature from the previous trough or peak to spawning.

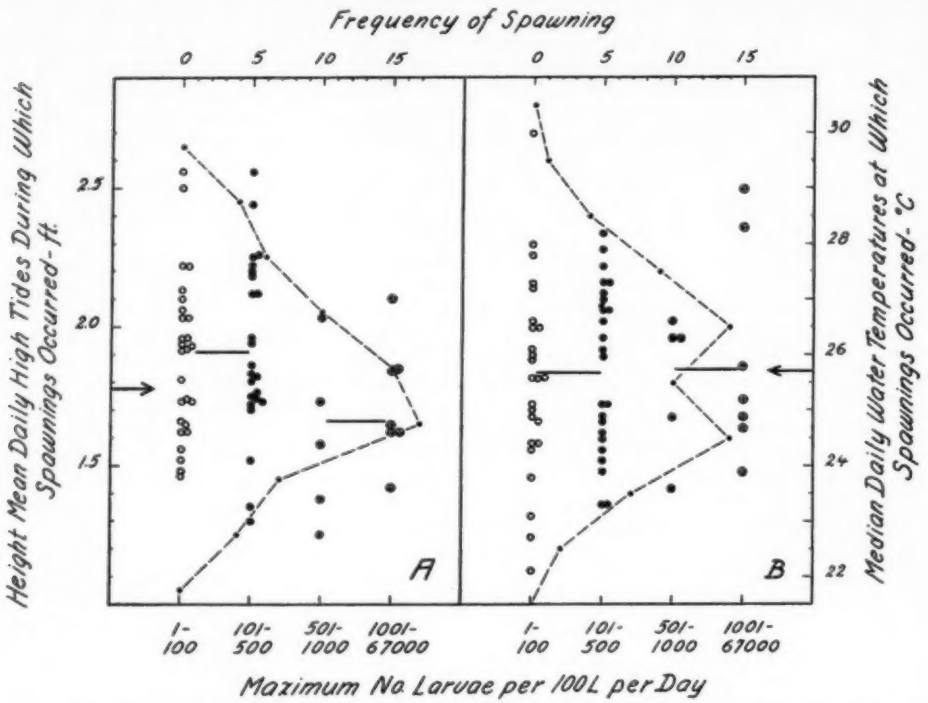


FIG. 29. Relation of spawning of swarms of larvae of *M. mercenaria* of different densities in Little Egg Harbor during summers of 1948–1951 to (A) height of mean daily high tides (roughly tidal amplitude) and (B) median daily water temperatures. See legend of Figure 28 for symbols of larval densities. Arrow extending from the y-axis in A designates the mean level of high tides at which all spawnings occurred (1.78 feet), tide levels throughout equated to 0 feet mean low water; bars in A indicate mean level of high tides at which spawnings of low-density swarms (1.91 feet) and high-density swarms (1.66 feet) occurred. Arrow extending from y-axis in B designates mean daily spawning temperature for all swarms (25.7°C), and the bars facing the arrow indicate mean daily spawning temperatures for the low- and high-density larval swarms. Mean minimum median summer water temperature for the four summers was 21.2°C, and mean maximum median summer water temperature was 29.1°C. Broken-line curves associate frequency of spawning and tidal amplitude (at 0.2 ft. intervals) in A, and temperature (1° intervals) in B.

over a median daily range of water temperatures of 22° to 30°C. but maximum frequency of spawning occurred within the thermal range of 24°–26°C. (Fig. 29b).

As suggested in Figures 24 through 27 and amplified in Figure 30, spawnings occurred more frequently and more high-density swarms of early-stage clam larvae appeared in the water during neap than during spring-tide periods. Mean amplitude of the four summer maximum tides was 2.88 feet, and mean amplitude of all tides of the four summers was 1.85 feet, based on a level at mean low water of zero. Mean amplitude of all tides during which spawnings occurred was 1.78 feet, and within a range of amplitudes of 1.25 to 2.56 feet (Fig. 29a). Low-density swarms were spawned at a mean tidal amplitude of 1.91 feet and high-density swarms at 1.66 feet. The frequency of spawning was maximal within a range of tidal amplitudes of 1.6 to 1.9 feet (Fig. 29a). Thus the total number of spawnings per day, as well as the density of swarms, increased from spring to neap tides, paralleling an increase in frequency of spawning with successive decreases of tidal amplitude to 1.6 feet.

Curves for median daily water temperature and mean daily tidal amplitude in Figures 24 through 27 frequently diverge and converge in a roughly symmetrical pattern. This correspondence suggests that during low tides

of neap periods, when minimal exchange occurred with the cooler ocean water, bay water temperatures rose above those of spring-tide periods. However, because of the distance of Station 4 from the inlet and general isolation of the mass of water at Station 4 (Fig. 16, 17, 20) from the inlet, the partial correspondence of these curves was coincidental. Study of the relationship of air temperature, water temperature, and tidal amplitude at Station 4 over periods of several summer months demonstrated a correlation between only air and water temperatures.

The cooling effect of ocean water was demonstrable in the lower portions of the central basin of Lower Little Egg Harbor (Fig. 20), but only during the late flood of high spring tides. Since maximum density of parents was met in the southern region of the central basin within the influence of cooling high spring tidal water, and maximum density of swarms of early veligers at Station 4 predominated during neap tides, it is probable that the higher water temperatures of neap tides triggered the spawning of these swarms (Fig. 30). The bulk of spawning probably took place at or close to the ebb slack period when maximal water temperatures prevailed, since larvae appeared in greatest number at Station 4 during late flood and early ebb tide, carried there by tidal transport from the area of maximum parental densities in the lower part of the central basin (Figs. 16, 17). Other factors than heat, such as kind and quantity of food (Thorson 1950), may possibly have contributed to spawning and should not be entirely discounted. Spawning at or just after low tide would be advantageous to the larvae, since they would be transported up-bay from the spawning area rather than down-bay toward the inlet, with possible loss to sea.

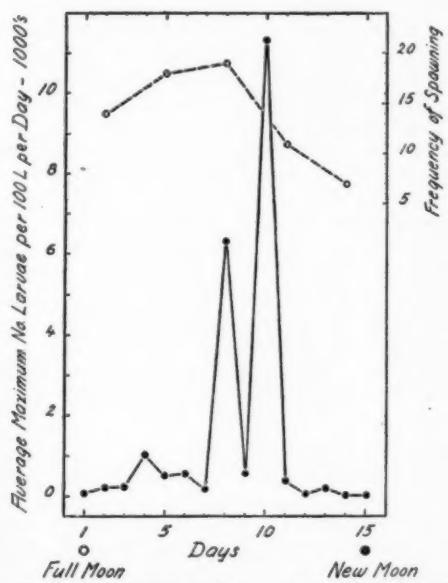


FIG. 30. Relation of frequency of spawning of *M. mercenaria* (3-day means; total of 67 spawnings in 4 summers) and spawning time, and relation of density of resulting larval swarms to full and new moon. All data are equated to a 15-day interval between full and new moon.

Landers (1954) reports that hard clams in intertidal areas in Narragansett Bay spawned earlier in the season than those in deeper water even though the prevailing temperature of the water mass over both was the same. In this ecological situation, warming of the exposed bottoms triggered release of gametes in the same manner as the higher water temperatures of neap-tide periods in Little Egg Harbor.

Data presented in Figures 24 through 27 permit approximation of the percentage of survival of clam larvae during planktonic existence. In Table IX are tabulated pertinent data on the ten densest swarms encountered during the four summers. In six of these swarms (marked by an asterisk), late umboned veliger and pediveliger larvae were found in the water toward the termination of planktonic existence. Approximate densities of early and late stages suggest a survival value (which probably also reflects an unknown degree of dispersion of the larvae) of roughly 2.6 per cent. These six swarms coincided with periods of little or no rainfall and medium to low tidal amplitudes (Fig. 15).

The four other swarms listed in Table IX (spawned on July 28–29, 1948; July 5, 1949; and July 11–12, 1951), however, completely disappeared from the water at Station 4 at an early age.

Loss of these larval swarms may be partially explained by flushing during periods of heavy rainfall and by high spring tidal exchanges, since in all four cases cited above larvae disappeared coincident with mean daily tidal amplitudes in excess of 2.1 feet (tidal exchanges exceeding 37 per cent) and rainfall ranging from 1.21 to 3.96 inches. This loss may be elucidated for the swarm of July 28–29, 1948. Assuming a mean mortality rate of larvae in Lower Little Egg Harbor of 97.4 per cent (Table IX) from predators and from a small degree of tidal exchange, 72 larvae per 100 liters of sea

Table IX

Rate of disappearance of clam veligers from the plankton of Little Egg Harbor.

Early Stages in Plankton			Late Stages in Plankton			Percentage survival of larvae
Date	Length range, μ	No. per 100 liters	Date	Length range, μ	No. per 100 liters	
July 30, '48	98–127	2,780	—	—	—	0
Aug. 10, '48	100–115	240	Aug. 18	180–196	20	8*
July 6, '49	98–140	2,100	—	—	—	0
June 25, '50	100–113	800	June 30	185	2	0.3*
July 7, '50	107–133	3,300	July 11	160–193	36	0.1*
July 19, '50	93–117	112	July 25	175–187	8	7*
July 12, '51	98–100	67,000	—	—	—	0
July 13, '51	104–114	34,000	—	—	—	0
July 23, '51	115–130	6,000	July 28	182	1	0.02*
July 24, '51	115–135	1,630	July 29	182–198	1	0.06*

Mean of * 3.6.
Over-all mean 1.6.

water of the original 2,780 larvae per 100 liters would reach setting size. During the last six days of planktonic existence of the swarm a total of 3.96 inches of rain fell, resulting in addition of rain water to the bay equivalent to about 10 per cent of the mean high-tide volume of Little Egg Harbor. Dilution by rainfall to this degree, assuming rather rapid mixing of fresh water with the bay water, would result in a decrease in density of the larvae which were ready to set from 72 larvae per 100 liters to about 65. But more important, during the latter portion of the planktonic existence of the swarm, tidal amplitudes increased from 1.4 to 2.7 feet, augmenting exchange from about 24 to 42 per cent. Although the data do not permit calculation of the quantity of loss of larvae by exchange, such large increases in exchange, compounded to a small degree by flushing, may well provide on explanation for much of larval loss.

It is evident from these observations that the strong tendeney of *M. mercenaria* to spawn more frequently and to produce densest swarms of larvae during neap tides in Little Egg Harbor has probably contributed to a retention and high degree of survival of the species in this bay.

Duration of Planktonic Stage of *M. mercenaria* in Little Egg Harbor. On June 25, 1950, a moderately dense though widespread single swarm of early straight-hinged veligers appeared on the late flood tide at Station 4 (Fig. 26). No other spawnings occurred in this area in June; it was therefore possible to follow growth of individuals in daily samples of the single swarm and determine duration of the planktonic stage. The swarm was accompanied by low to moderate tidal amplitudes and very little rainfall (Fig. 15: 1950), which probably accounts for survival of a small proportion of the population to setting size. In typical fashion, the density of larvae in the daily plankton samples decreased rapidly with time (Fig. 31) from a maximum of about 800 to a minimum of two larvae per 100 liters. Growth rate, indicated by the length of the most abundant size group of larvae in each plankton sample, was rapid. The largest larvae reached a length of 185 μ in seven days from the time of fertilization of ova, paralleling the most rapid rates expressed by larvae cultured in our laboratory and almost those in that of Loosanoff and associates (1951b), who have obtained a minimum setting time of six days. Extrapolation from the growth curve in Figure 31 shows that these larvae grew to a setting size of approximately 200 μ in eight days in native waters. Median daily water temperatures ranged from 23.4° to 26.2°C. and mean daily salinities from 30.4 to 31.4 °/oo during the larval period (Fig. 31).

The length of abundant size groups of larvae was determined for most samples collected for the remainder of the summer of 1950. Growth curves represented by these parallel rather

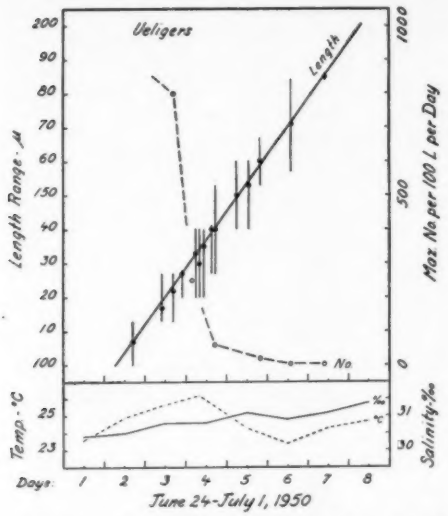


FIG. 31. Rate of growth and of disappearance of veligers of *M. mercenaria* in a large, single, widespread swarm appearing in the central basin of Lower Little Egg Harbor (Stations 2–4) on June 25 (spawned June 24), 1950 (compare with Figure 26), showing length range in vertical lines; and most abundant size group of larvae in solid circles. Connected open circles represent maximum densities of swarm from day to day. Accompanying temperature (median daily) and salinities (average of surface and bottom) are given in bottom figure.

closely the growth curve of the population spawned on June 24 (Fig. 26). This same population was accompanied by relatively high water temperatures, which accelerated larval growth rates. In contrast, the population spawned in late July of the same summer was confronted with rapidly dropping temperatures which slowed growth rate and prolonged the veliger period. These growth curves, as did those of Loosanoff et al. (1951b) for larvae grown in the laboratory, approximate straight lines.

HORIZONTAL AND VERTICAL DISTRIBUTION OF CLAM LARVAE. Older stages of planktonic larvae of a few species of estuarine animals—the eastern oyster *Crassostrea virginica* (Nelson and Perkins, 1931; Carriker, 1951; Pritchard, 1951; Manning and Whaley, 1954) and the barnacles *Balanus improvisus, B. crenatus,* and *B. balanoides* (Bousfield, 1955)—tend to concentrate in lower, more saline strata of sea water and are thereby transported passively up the estuary in the net nontidal upstream flow (Pritchard 1952). Hulburt (1957) has shown that both larvae and adults of the planktonic shrimp *Neomysis americana* behave in a similar manner. In the case of the oyster and the barnacles, larvae may thus set some distance up the estuary from spawning parents. The eastern oyster is the only mollusk in which ecological studies of this kind have been reported; our investigations in Little Egg Harbor provided an opportunity to extend observations of vertical and horizontal movements of mollusk veligers to the hard clam.

The most striking feature of the horizontal distribution of veligers of *M. mercenaria* in Little Egg Harbor was its unevenness. Following occasional widespread spawning of adults during the early part of the summer, larvae were encountered at all stages of the tide throughout Lower Little Egg Harbor and the lower reaches of tributary tidal creeks. Maximum densities were found in the region around Stations 3, 4, and 8. During the summer of 1948 a maximum of 2,780 early straight-hinged larvae per 100 liters of sea water was taken there; in 1949 this maximum was 3,800; in 1950, 3,500; and in 1951, 67,200. Densities decreased progressively, though quite irregularly, toward Manahawkin Bay, Tuckerton Creek, and Beach Haven-Little Egg Inlets.

Low larval densities near inlets resulted in part from the diluting effect of tidal exchange. That larvae from the lower areas of Lower Little Egg Harbor were transported out of the bay was demonstrated on August 8, 1950, when we collected a vertical series of plankton samples at hourly intervals from late ebb tide through the subsequent flooding tide to flood slack at Station 10 in the inlet. During this 7.5-hour interval, the ranges of mean vertical water temperatures and salinities were 0.7°C. and 0.5 °/oo respectively, suggesting the homogeneity of the water mass. The first high tide of the day was 0.35 feet below mean high water (Table VI), and about 30 per cent of the bay water emptied into the ocean during the ebbing tide. During late ebb and early flood tide an average of 0.2 late umboned veligers was present per 100 liters, whereas no larvae were encountered during mid and late flood tide. The previous day at mid ebb tide an average of 35 late umboned clam larvae per 100 liters was taken in a vertical series of plankton samples pumped at Station 4. A second series at the same station and at the same phase of the tide on August 9 disclosed that the average density of late umboned veligers had dropped to 21.

After limited localized spawning of small segments of the clam population in the bay, larvae appeared in small swarms of variable densities and could be detected as they passed Station 4 in the tidal flow. Individual swarms could often be followed from day to day at this station by sampling at approximately the same phase of the tidal cycle.

In the laboratory a small rise in water temperature will stimulate a few male hard clams to emit spermatozoa when they are physiologically ready to spawn, and the double stimulus of temperature and sperm will bring on spawning of other similar individuals in the same water, resulting in a dense suspension of gametes. Although not actually observed in the field, this behavior can be attributed to populations in the bay to explain the grouping of larvae in swarms. Thermal stimulation of a few males in the bottom will induce release of sperm, which in turn will cause other males in the close vicinity to spawn; in a few minutes females will commence discharging ova. Soon tidal flow will spread over the bottom a cloud of gametes which will be transported through the distance of the tidal excursion. If the few initiating male spawners are widespread across the main axis of tidal circulation in the bay, and clams are generally ready to release gametes a mass spawning of baywide dimensions will occur. During the early part of the season when most abundant spawnings took place, we did encounter such widely distributed swarms of larvae of relatively uniform size in Lower Little Egg Harbor. Later in the summer, spawning in the laboratory was obtained with more difficulty and in limited quantities, and correspondingly smaller localized larval swarms occurred sporadically in the bay. Thorson (1950) states that it may be considered a biological rule that among invertebrates which shed their eggs and sperm freely in the water, the males are the first to spawn and thus stimulate the females to shed their eggs; these, having been shed directly into a suspension of sperm of their own species, have especially good chances of being fertilized.

We collected and studied a total of 97 vertical series of plankton samples over the period of four summers in Little Egg Harbor and tributaries, each series consisting of from three to six vertical samples equally spaced from surface to bottom. Most samples were pumped at Station 1 in Ezras Creek and at Station 4 in the open bay (Fig. 14). In this way it was possible to contrast distribution and movements of veligers in a shallow, narrow tidal creek with that in the open bay. Data on distribution were analyzed in three ways:

I. The first analysis is based on a study of plots in the form of polygons of representative vertical series of veliger densities and accompanying physical data (Figs. 32–35). Of

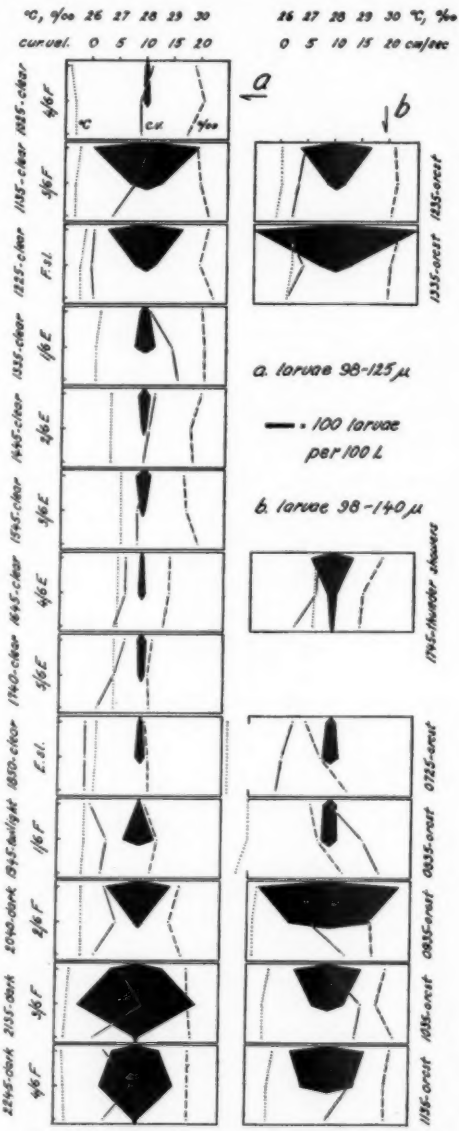


FIG. 32. Vertical distribution of straight-hinged veligers of *M. mercenaria* at approximately hourly intervals through the cycle of two separate tides at Station 1 in the middle of Ezras Creek, Little

the several series of vertical plankton samples pumped through the cycle of a tide at Station 1, those on July 26 (Fig. 32a) and July 27, 1948 (Fig. 32b), contained maximum concentrations of straight-hinged veligers and are representative of veliger distribution and movements in the creek. Characteristically, very few umboned larvae were found there. Maximum densities appeared during the late flood tide, the higher evening tide of July 26 bringing high larval concentrations earlier in the tidal cycle than the lower morning tide, as water from the lower portion of the central basin entered the creek. Densities decreased rapidly in the course of the ebbing tide, presumably because of inroads of bottom-dwelling filter-feeding predators. The polygons suggest that during daylight hours and while creek water was ebbing and flooding swiftly, maximal larval densities remained near the surface. As current velocity decelerated during daylight hours, these dropped to about mid-depth, only to rise surfaceward again as tidal movement accelerated. Early veliger stages of *Crassostrea virginica* likewise ascend into surface layers concomitant with maximal current velocities of flood and ebb tides (Carriker, 1951). I would like to suggest that turbulence resulting from the movement of sea water in the irregular creek channel was the stimulus which promoted their surfaceward climb. As here employed, turbulence refers to the state in which random motion of smaller or larger masses of sea water is superimposed upon the general pattern of flow (Sverdrup et al., 1942). Soon after darkness fell (3/6 and 4/6 F. Fig. 32a), the zone of maximum larval density shifted to midstream, and a significant number of larvae were collected near the bottom in spite of strong currents and in contrast to the surface position of maximal densities at similar phases of the tide during daylight hours on July 27 (Fig. 32b). This distributional pattern suggests that at night the larvae, no longer attracted toward the surface by light, assumed a broader vertical distribution and in addition sought the zone of maximum current velocity (turbulence?) at midstream. In a total of thirty-four vertical series of plankton samples collected in the creek during daylight hours, larvae were encountered near the bottom in only three, and in these only in negligible numbers. No explanation is available for this, except for one (1745, July 27, Fig. 32b) taken shortly after a severe thunderstorm in which conditions accompanying the storm may have sent a few of the veligers bottomward. In contrast, Quayle (1952), in a preliminary study of vertical distribution of veligers of the clam *Venerupis pullastra* in Fairlie Channel, Scotland, found the larvae widely distributed in depth during the day but concentrated near the surface at night. In his study of the distribution of the veliger larvae of *Ostrea edulis* in the Oosterschelde, Holland, Korringa (1940) observed that vertical distribution of the larvae appeared to be uninfluenced by light, water temperature, wind and wave action, and current velocity, larvae remaining more or less uniformly distributed vertically under these conditions. And Carriker (1951) and Manning and Whaley (1954) report that older veliger stages of *Crassostrea virginica* occur in maximal densities at progressively greater depths as they approach the pediveliger stage. Since the larvae of *M. mercenaria* appear able to swim faster than the vertical water currents and to some degree maintain a vertical position, it is probable they are also concentrated horizontally by typical water movements (Pomeroy et al., 1956; Ragotzkie et al., 1953).

At Station 4 in Little Egg Harbor maximal larval densities were frequently found during the late flood and early ebb tide (Figs. 33, 34). As in the creek, during daylight hours larvae were generally absent near the bottom. Two exceptions to this were met in a total of 53 vertical series of plankton samples pumped at all stations in the central basin of the bay.

Egg Harbor, N. J.: left, 1025 to 2245, July 26; right, 0725 to 1745, July 27. Hour indicates the mid point in sampling interval for each vertical series. The study of July 26 was extended into the evening to include effect of darkness on larval distribution; collecting at 1745 in the study of July 27 was done during a thunderstorm. For each vertical series there are shown the density of veligers at surface, middle, and bottom and accompanying temperature, salinity, and current velocity of the water as well as approximate quantity of light (degree of cloudiness of the sky, and whether day or night). The breeze on July 26 was moderate (Beaufort Scale of Winds No. 4) throughout the day, and the interval of time for collecting all data at each vertical series of three strata ranged from seven to ten minutes; on July 27 the breeze ranged from calm to light (Beaufort No. 0–1), and interval of time for collecting data at each vertical series ranged from six to nine minutes. The depth of water at the point of sampling on July 26 ranged from 1.3 m. at ebb slack to 1.8 m. at flood slack; and on July 27, from 1.1 m. to 1.7 m. Data for July 27 are plotted so that stages of the tide coincide with those of July 26; the vertical series for 1/6-3/6 E were omitted on July 27.

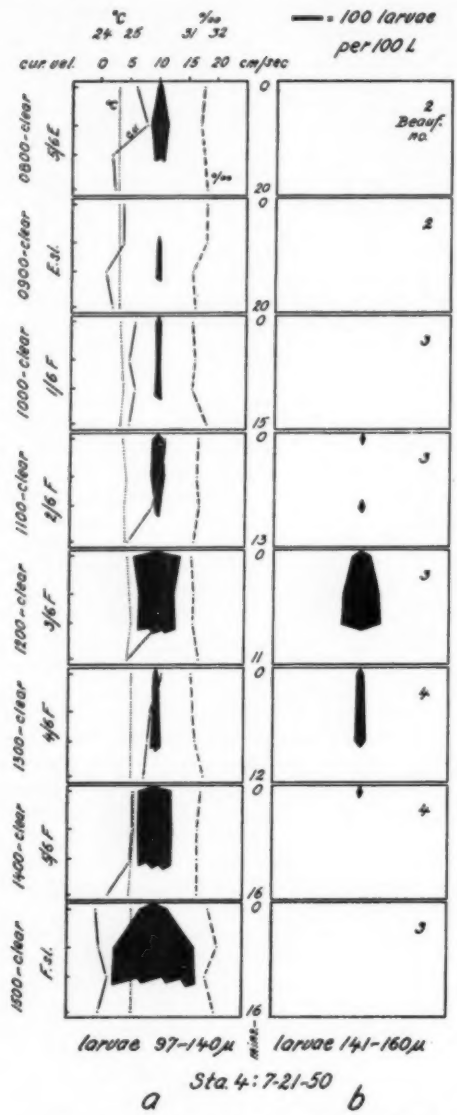


FIG. 33. Vertical distribution of straight-hinged (left) and umboned (right) veligers of *M. mercenaria* at hourly intervals through two-thirds of a tidal cycle in Little Egg Harbor, N. J., during daylight. Larvae were sampled at four equally spaced strata from surface to bottom. Accompanying data on water temperature, salinity, current velocity, condition of the sky, intensity of the wind, and duration of sampling interval for each vertical series (between columns of rectangles) are shown. The depth of water at the sampling point ranged from 2.3 m. at ebb slack to 2.9 m. at flood slack tide. Densities of straight-hinged veligers in bottom strata were not recorded because of excessive quantity of sediment in the plankton samples.

No interpretation is available for the first (Fig. 34: 5/6 F–1245, 8–4–50, Sta. 4); the second, found at Station 8 (Fig. 34, top rectangles), may be due to location of the station in a region characterized by a complex pattern of currents. Main larval concentrations in the bay, in contrast to those in the creek, occurred principally somewhere in the middle of the vertical column regardless of the stage of the tide. Possibly the lower turbulence in the bay than in the creek may account for this. The apparent exception noted in the top set of rectangles in Figure 34 may be explained by the fact that these samples were collected in a water mass which had just passed through channels among islands leading to the central basin (Fig. 14), and the larvae in these channels behaved as they would have in the creek. During hours of darkness in the bay, veligers were generally distributed rather uniformly from surface to bottom (Fig. 35). In a total of seven vertical series of plankton samples pumped there at night, a single exception was noted (top right rectangle, Fig. 35). Whether this is attributable to sampling during slack water rather than during periods of tidal flow is not known. Because of the vertical uniformity of temperature and salinity in the bay and in the creek, no effort was made to associate vertical distribution of larvae with these factors.

II. In the second analysis of the data, larval density at each stratum was converted to percentage of total larvae in its respective vertical series, and vertical series were grouped according to location, early- and late-stage larvae, stage of the tidal cycle, wind velocity, and relative intensity of light. The mean percentage of larvae in each vertical stratum was then calculated for each group, and these percentage means were plotted for ease of interpretation. Although the significance of the results obtained by this kind of analysis may rightly be questioned, it is worth noting that the results tend to support those of the first analysis (Figs. 32–35) and to suggest additional trends. Whereas in the creek larvae rose surfaceward on the early flood and early ebb and dropped to the middle again on the late flood and ebb, these relationships were less clear in the bay: there larvae appeared to rise on the early flood, straight-hinged ahead of umboned larvae, then to sink slowly to the middle stratum through-

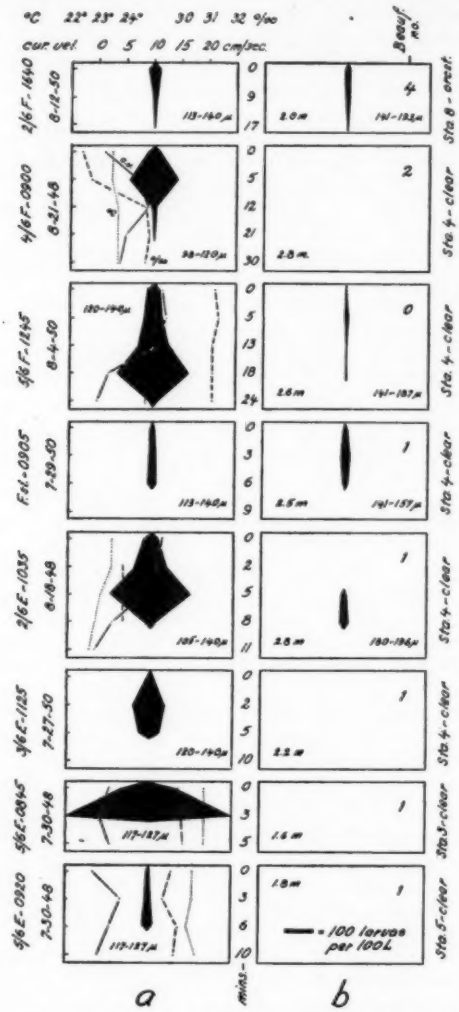


FIG. 34. Vertical distribution of straight-hinged (left) and umboned (right) veligers of *M. mercenaria* in a series of isolated vertical studies made at different stations in Little Egg Harbor, N. J., during daylight and arranged in the natural sequence of the stages of the tidal cycle. Larvae were sampled at equal intervals from surface to bottom. Each vertical series is identified by station number and hour and date the sample was taken. Where available, accompanying data on water temperature, salinity, current velocity, condition of the sky, intensity of the wind, duration of sampling interval for each vertical series, and range of length of larvae are indicated. (The density of straight-hinged veligers in the "5/6 E–0845" vertical series is approximately five times that indicated by the density scale). The depth of water at sampling time is given in meters at the lower left in each of the righthand rectangles.

out the remainder of the flood and ebb tide. Further, as wind velocity increased in the bay during daylight hours, veligers seemed to rise vertically, straight-hinged preceding umboned stages. This relationship, if real, may again indicate a response of larvae to augmented turbulence of the water, in this instance brought about by wave action. As light intensity diminished in the bay, larvae appeared to move surfaceward, then spread somewhat uniformly throughout the vertical column from surface to bottom. In the creek, possibly because of greater turbulence there, maximal densities fluctuated between midstream and surface throughout day and night according to the stage of the tidal cycle, and only a few larvae moved near the bottom during darkness.

III. The third treatment of the data was by the analysis of variance of larval densities. To perform an analysis of variance we assume a linear model of the form log (y + 1) = (factor 1) + (factor 2) + . . . + (factor k), where y = the number of veligers per 100 liters of sea water. This transformation was made in the hope that log (y + 1) would provide a scale on which the various environmental effects would be additive, and the deviations from the model would have approximately a normal distribution. Similar transformations have been made by Winsor and Clark (1940).

The following factors were considered in the equation used in analyzing the data for both the straight-hinged and umboned larvae:

1. Stage of the tidal cycle: 12-hour intervals.
2. Time of day: 0400–0700, 0700–1700, 1700–2000, 2000–0400.
3. Depth of vertical samples: equally spaced between surface and bottom; in deeper water: surface, upper middle, middle, lower middle; and in shallower water: surface, upper middle, lower middle (bottom samples were excluded since so few larvae were taken there).
4. Wind velocity: calm, light, moderate, strong (Beaufort Nos. 0–1, 2–3, 4, 5).
5. Light: degree of cloudiness of sky (one-third, two-thirds, and fully overcast; twilight); and darkness (nighttime).
6. Location of Stations 1, 3, 4, 5, and 8.
7. Interactions of (a) relative intensity of light, (b) relative wind velocity, (c) stage of the tidal cycle, and (d) day and night, with depth.
8. Current velocity.

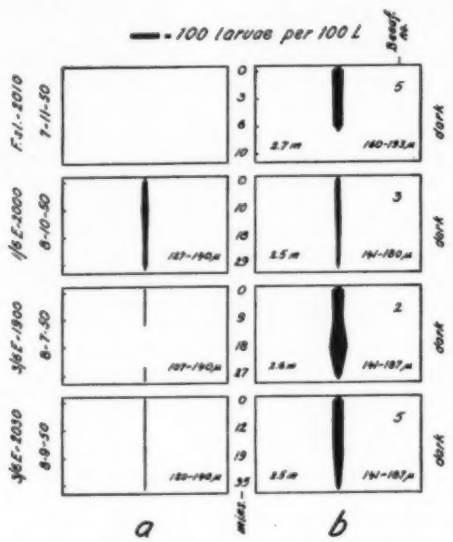


FIG. 35. Vertical distribution of straight-hinged (left) and umboned (right) veligers of *M. mercenaria* in a series of isolated vertical studies made at Station 4 in Little Egg Harbor, N. J., at night. Larvae were sampled at equal intervals from surface to bottom. The intensity of the wind, duration of sampling interval for each vertical series, range of length of larvae, and depth of water at sampling time are indicated.

The basic assumption drawn in formulation of the equation, that effect of environmental factors is approximately additive, is verified by the fact that no interactions were found to be significant. The assumption of normality of the deviations from the model was investigated by the *chi*-squared test of goodness of fit. For the early stage larvae, *chi*-squared with 11 degrees of freedom was 8.74 which indicated close agreement with the hypothesized normal distribution. However this was not the case with the late-stage larvae. A histogram of the deviations showed a long tail on the right—a result of the fact that many of the catches were zero in number. Many investigations have been made of the effect of nonnormality on inferences made from the analysis of variance. Scheffé (1959) shows that inferences about means are not seriously upset by nonnormality. Fortunately in this investigation no cases of borderline significance were encountered. Thus it is unlikely that more than the expected number of errors in inference will be made. It should also be emphasized that ecological relationships which are not statistically significant according to this statistical analysis may in reality be significant, but obscured by treatment of the data or insufficient information. However, any that are found are not likely to be artifacts. It is for this reason that interpretation of plots of hour-to-hour distribution of larvae with reference to accompanying environmental factors was presented in the first analysis.

Results of the statistical analysis may be summarized as follows. Appearance of maximal larval densities in the creek during the late flooding tide is statistically significant ($p < .01$) and confirms what is shown in Figure 32; the water which enters the creek at this period of the tidal cycle bears high concentrations of veligers from the lower portion of the central basin of the bay. Also significant, though unexplained, is the increase in density of larvae in the creek as the relative intensity of light decreased during the day ($p < .01$). Taken as a whole, the analysis suggests that the stage of the tide, current velocity, relative quantity of light during daylight hours, and time of day had no significant influence on the vertical distribution, except for the bottom stratum, of the larvae in the creek. However, the detailed hour-to-hour study reported in Figure 32 shows trends not found significant by this method of analysis.

Appearance of maximal concentrations of straight-hinged veligers in the bay at Station 4 during the late flood and early ebb is statistically significant ($p < .01$); in contrast, umboned larvae were more or less uniformly distributed throughout the tidal cycle (Fig. 36). This observation finds support in the fact that the maximum density of spawning parents was located in the lower part of the central basin, and it suggests that spawning probably occurred during late ebb or early flood. It further suggests that early-stage larvae tended to remain in the original swarms resulting from mass spawning by parents, and in time, as a result of the hydrographic mixing processes, late-stage larvae became widely and rather uniformly dispersed and no longer exhibited noticeable peaks of concentration within the excursion of a tidal cycle

The linear effect of degree of overcast of the sky on horizontal distribution of larvae in the bay was not significant. Yet the time of day provided a large and statistically significant correlation with larval density (Table X) ($p < .01$). Maximum densities (expressed as geometric means) of straight-hinged veligers were encountered in the daylight period of 0700–1700, whereas most umboned larvae were

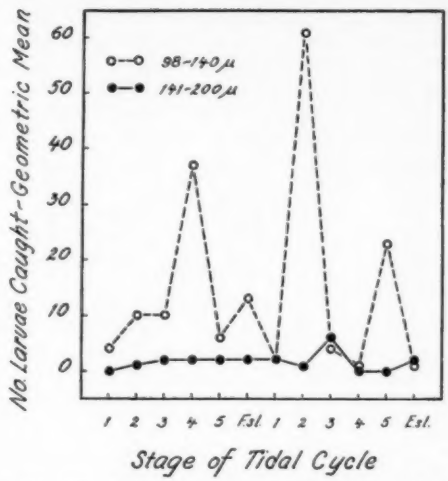


FIG. 36. Geometric means of the total number of straight-hinged and umboned veliger larvae of *M. mercenaria* caught at all depths—day and night—at each stage of the tidal cycle during the study in Little Egg Harbor, N. J.

found during hours of darkness. No data are available which indicate that bivalve veligers may spend a part of the twenty-four-hour period at rest on the bottom, while Gaarder and Bjerkan (1934) have shown that veligers of *Ostrea edulis* raised in Norwegian oyster pools must remain entirely off the bottom because of the lack of oxygen and presence of high concentrations of hydrogen sulfide there.

Table X

Geometric mean of number of veligers of *M. mercenaria* per 100 liters collected in Little Egg Harbor, N. J., during different periods of the day.

Hours	No. of samples	Straight-hinged veligers	Umboned veligers
0400–0700.....	3	4	0
0700–1700.....	144	12	1
1700–2000.....	21	5	7
2000–0400.....	9	1	9

And Dew and Wood (1955) found bivalve larvae (chiefly *Crassostrea commercialis*) more dense in statistically significant quantities in both surface and deep tows at night than during the day in Sydney Harbor, Australia. It would seem that differences in vertical distribution of larvae in both Little Egg Harbor and in Sydney Harbor during day and night and sampling at constant vertical strata may account for this apparent diurnal density variation.

Increasing wind velocity had no apparent linear influence on density of straight-hinged veligers at Station 4, but there was a statistically significant increase in the density of umboned larvae with increase of wind velocity ($p < .05$). This apparently anomalous circumstance is explained by the fact that prevailing winds, and particularly moderate to strong ones, in the Little Egg Harbor area during the summer moved up the bay from the southwest (Fig. 14), forcing masses of water bearing umboned larvae toward Station 4. The difference in distribution between early- and late-stage larvae may suggest that older stages tend to be transported down the bay as they develop. If this hypothesis is valid, a response of veligers to migrate surfaceward under the double stimulus of turbulence and light and the presence of even very slight nontidal surface water seaward would provide a mechanism by means of which veligers could be transported seaward as they approach the pediveliger stage. The daylight period is longer than nighttime in the summer in Little Egg Harbor, and winds are present often over much of the twenty-four-hour period. The vertical uniformity of salinity and water temperature in the bay precluded a study of the influence of these factors on vertical distribution, but does not exclude the possible presence of weak nontidal circulation (Pritchard, 1952). These problems merit further study.

The degree of cloudiness of the sky during the day had no statistically significant effect on vertical distribution of the larvae.

An analysis of the over-all mean vertical distribution of veligers in vertical series collected in the bay under all conditions (geometric means, Table XI) suggests that maximum density occurred in the middle zone for the straight-hinged veligers, and that umboned larvae were rather uniformly distributed in the vertical column except for the bottom stratum. Even though the geometric mean of number of veligers in the bottom stratum of both early and late stages is 1, most samples did not contain larvae of either stage. However, the vertical variation of density of straight-hinged larvae shown in the table is not statistically significant. This is surprising and may be accounted for by the facts that (a) the number of samples at the middle stratum is relatively small; and (b) five of the eleven samples

Table XI

Geometric mean of number of veligers of *M. mercenaria* per 100 liters collected in different vertical strata, day and night, in Little Egg Harbor, N. J.

Depth	No. samples at each depth	Straight-hinged veligers	Umboned veligers
Surface	58	8	2
Upper middle	54	11	2
Middle	11	33	1
Lower middle	54	7	2
Bottom	58	1	1

were taken during 4/6 flood and 2/6 ebb tide, which were tidal stages of maximum larval densities, and ten of the eleven samples were collected during the time interval 0700–1700, which was also a period of high larval densities. It should be noted that plots of representative vertical series in Figure 34 likewise point to maximum larval concentrations somewhere in the middle stratum. The stage of the tide, current velocity, relative intensity of light during the day, and wind velocity had no significant influence on the vertical distribution of the larvae (exclusive of the bottom) in the bay. But the time of day (and concomitantly, the effect of day and night) did appear to have some effect on the vertical distribution of umboned veligers in the bay (Table XII). They appeared to remain rather uniformly distributed above the bottom during daylight hours and to move bottomward during twilight and hours of darkness. This supports the polygonal plots of distribution in Figure 35. Straight-hinged veligers, as also shown in Table X, tended to concentrate near the middle stratum, at least during daylight and early evening hours. These observations would indicate that the larvae are able to control their vertical distribution to some degree, possibly seeking a certain range of light intensity during the day, and are more than passive buoyant particles in the water (Turner, 1953).

In summary, these three analyses of vertical distribution of veligers of *M. mercenaria* suggest the following possible trends: (1) Veligers seem to maintain themselves in the water throughout the planktonic existence, generally away from the bottom and bottom-dwelling predators during daylight hours; they are more broadly distributed (descending bottomward, especially the umboned stages) during hours of darkness. This raises the question

Table XII

Geometric mean of number of veligers of *M. mercenaria* per 100 liters collected in different vertical strata during daylight hours (0700–1700) and during evening hours (1700–2000; 2000–0400).

Depth	No. samples at each depth	Straight-hinged veligers	Umboned veligers
0700–1700			
Surface	47	10	1
Upper middle	44	15	1
Middle	10	35	1
Lower middle	43	10	2
Bottom	47	1	0
1700–2000			
Surface	7	4	6
Upper middle	7	6	8
Lower middle	7	4	9
Bottom	7	1	2
2000–0400			
Surface	3	1	9
Upper middle	3	0	11
Lower middle	3	1	8
Bottom	3	1	2

whether pediveligers may set most frequently at night. The major concentration of straight-hinged larvae in the middle strata during daylight hours may indicate that they are attracted to an optimum light intensity in that zone, whereas at night this stimulus disappears and larvae then mix more uniformly throughout the column. (2) Veligers may be stimulated to rise in the water column under the stimulus of turbulence created by tidal currents and wave action. This behavior would make possible differential horizontal migrations in the presence of net nontidal estuarine circulation. Laboratory experiments of Turner and George (1955), which showed that hard-clam veligers are blocked in their vertical movement through a gradient of salinity by the 15 °/oo stratum, suggest a means by which larvae may move up the estuary in the presence of low surface salinities. Where a halicline at the 15 °/oo level coincided with the vertical level of no net motion of horizontal estuarine circulation, it would appear that larvae

would remain entirely within the bottom net up-estuary flow.

Thorson (1950) emphasizes the point that young pelagic larvae of marine invertebrates (except those whose adults live in the intertidal zone or near the surface) are positively phototactic, while old larvae approaching metamorphosis are negatively phototactic. Thus larvae spend their pelagic existence in the free water masses near the illuminated-phytoplankton-producing strata near the surface, while metamorphosing larvae, because of their photonegativity, are forced toward the bottom to the substratum, where they will settle. Thorson's observations suggest that the vertical distribution of veligers of *M. mercenaria* may be explained in part as a response to distribution of food organisms; at present there is no information on this interesting point.

The field studies on distribution reported here are of a preliminary nature. Although we collected many samples, vagaries of the weather, erratic temporal and horizontal distribution of the larvae in the bay, and short duration and precipitous loss of veligers during the planktonic existence all militated against a more productive sampling design of larval populations. In addition to providing some suggestions on larval ecology of *M. mercenaria* in the field, we hope that these studies will stimulate a more rigorous approach to such studies in the future. With the assistance of modern technology it should be possible to devise and employ a series of multiple plankton samplers by means of which larval populations and pertinent environmental factors could be sampled simultaneously at a number of vertical strata in several different stations along the long axis of an estuary at half-hourly intervals during the cycle of many tides when dense larval swarms are in the water. Comparative investigations in a variety of estuaries and embayments would contribute additional valuable information.

Pediveliger and Byssal Plantigrade Stages in the Laboratory

Pediveligers are extremely active. After the foot is formed, they display it as they swim, extending it periodically between the valves, possibly in an attempt to contact the substratum. Larvae ranging in length from 170 to 220 μ were observed exhibiting this behavior. On the bottom pediveligers creep smoothly over the substratum for a while, moving back and forth in what looks like a searching pattern, then suddenly extend the velum, swim off to a different site, settle to the bottom, and continue crawling in a zigzag trail. Alternation of swimming and creeping may represent the animal's search for favorable setting surfaces, and the capacity to swim during this stage extends widely the territory which it can examine (Turner, 1953; Turner and George, 1955).

RESPONSE TO CONTACT. In 1951 we experimented to determine the attraction of a number of selected solid substrates and sediments of varying particle size to setting pediveligers. The following surfaces were placed on the plunger plate and on the bottom of mass larval cultures as larvae approached the pediveliger stage: (a) clean glass (small watch glasses); (b) clean, smooth, bleached oyster shell (*Crassostrea virginica*) and clam shell (*Mercenaria mercenaria*); (c) glass surfaces (microscope slides) dipped in melted paraffin and sprinkled with varying quantities of fine sand; (d) clean, bleached oyster and clam shell deeply excavated by the boring sponge *Cliona;* (e) deeply pitted leached slag; (f) small watch crystals in which detritus (microscopic fecal pellets, unconsolidated organic fragments, and some silt) had accumulated; (g) small watch crystals filled to varying depths with fine bay sand washed and screened through Screen No. 60; (h) similar watch crystals containing coarser sands. To find newly set plantigrades in fine sediment with a minimum of fatigue we used bright direct sunlight and a binocular magnification of about 25X.

No pediveligers affixed on surfaces placed on the plunger jar plates, probably because of the slow vertical movement of the plates. All setting took place on or near the bottom of culture jars. A few clams attached to the under side of some slightly elevated shells, but by far the majority affixed to the upper surfaces of materials provided. This observation supports Quayle's (1952) statement that most noncementing species prefer to settle on upper surfaces. Although setting in significant densities occurred in dishes partly filled with sand and in pits in slag and shell, densest sets were found on surfaces covered with detritus. In a watch crystal coated with a thin layer of paraffin and sand, the only detritus present at the time of setting had been incorporated in a worm case by a polychaete which had set in the dish; all clams in the dish attached to the worm case. Shells on the bottom of one of the mass clam cultures were coated with varying

thicknesses of detritus; attached clams were most dense on shells bearing the thicker layers of detritus. On pitted slag and shell, pediveligers accumulated most densely in the pits, where sediment also accumulated most deeply, trapped by the quiet pockets of water in the pits. In many pits pediveligers were packed so densely that no others could have entered. It was apparently not so much depressions that attracted pediveligers as sediment: slag coated with detritus bristled with clams, which were equally dense in detritus-filled pits and aggregations of detritus on open surfaces of the slag. Turner and George (1955) report that young byssal plantigrades of *M. mercenaria* in their laboratory likewise affixed in clumps of detritus and suggest that young clams prefer an upright position facilitated by the presence of detritus. Quayle (1952) found that young of *Venerupis pullastra* also settle on surfaces layered with detritus.

Where the depth of sediment did not greatly exceed the length of byssus and plantigrade combined, plantigrades affixed the byssus to the underlying solid surface and then remained suspended in the overlying sediment near the surface. In deeper sediment the majority affixed to glass around the inner surface of the dish and slightly below the surface level of the sediment; a minority, to grains of sand in the sediment. Plantigrades reared in the absence of sediment on the bottom of the container soon become coated with accumulating detritus, while those burrowing within sediment remained clean and smooth as a result of movement through the substrate.

These observations suggested that byssal plantigrades possess a strong thigmokinetic response. This hypothesis was supported by further preliminary experiments. With no exception those placed in clean dishes devoid of sediment attempted with continuing persistence to "dig" the distal tip of the foot into the glass. In a second experiment a population of plantigrades 0.2 to 0.4 mm. in length was maintained without sediment for eight days. These clams were then transferred to a petri dish in which the bottom was covered with a thin, uneven layer of clean sand; the sand particles ranged in diameter from 10 to 120 μ. The bivalves soon began crawling actively and randomly about, snaillike, through the shallow sand, and more vigorously than on a clean glass bottom alone, until deeper sand was encountered. They continued creeping through sediment until the valves were well covered, then stopped crawling and attached. The same plantigrades, when placed in a dish with larger particles of sand (about 400 μ in diameter), crawled less actively and attached less readily than in the fine sand, suggesting that a sediment of particle size smaller than the plantigrade is more suitable than larger particles for affixation. In this connection de Blok and Geelen (1958) demonstrated that newly metamorphosed *Mytilus edulis* show a decided preference for threadlike materials as attachment substrates for the byssus, and with increase in size the mussels select threads of larger diameter. Plantigrades which had attached to firm substratum under shallow sediment, when uncovered by removal of sediment by gentle flushing with a stream of water, quickly released the byssus and began creeping.

In the absence of suitable sources of contact stimulation plantigrades aggregated and received contact stimulation from each other. In a third experiment we placed some 50,000 plantigrades ranging in length from 0.2 to 0.5 mm. in a clean finger bowl in shallow sea water. Within an hour the majority had gathered in grapelike clusters and attached to the bottom and to each other. Hunter (1949) reports a similar strong contact response in byssal plantigrades of the rock-boring bivalve *Hiatella;* Morton (1960) makes the same report of adult *Lasaea rubra.*

These observations and experiments suggest that plantigrades of *M. mercenaria* set most abundantly in fine sediment containing appreciable quantities of organic detritus. It was not possible to determine the relative attractiveness to plantigrades of graded series of grain sizes of different kinds of organic and inorganic sediment. This should prove a profitable avenue for future research.

RESPONSE TO LIGHT. Whenever light reflected from the northern sky was directed on plantigrades (lengths ranging from 0.2 to 1.3 mm.) in a dish without sediment in the laboratory during clear days, the animals became restless, and many started crawling randomly about, their behavior suggesting a strong negative photokinetic response at this illumination. Measuring external dimensions of these young clams under a binocular microscope was therefore difficult because the light stimulated locomotion. On the average, locomotory activity was stimulated about 30 seconds after light was directed on the animal. Plantigrades buried under fine clean sand also became active when light was directed on them, the most exposed

animals expressing the most activity. Plantigrades buried more deeply in sediment (particularly in dark grains) were affected less or not at all by the light.

During the summer of 1956 we were able to examine this response more quantitatively. About 500 very active, rapidly growing plantigrades which ranged in length from 0.4 to 1.3 mm. were placed several layers deep within a circle two cm. in diameter in the center of each of two 6 cm. petri dishes, in sea water (of salinity 35.0 °/oo, temperature 27°C.) 5 mm. deep. One dish, the control, was set aside in the dim light of the laboratory (heavily overcast day, with no laboratory lights on). The experimental dish was placed on the glass stage of a binocular microscope and a beam of light from a microscope lamp, cooled through a water cell, was directed under the clams by a flat substage mirror; 700 foot-candles struck the animals. A mask with an opening 5.5 cm. in diameter under the petri dish provided a narrow shaded zone around the edge of the dish. Light was most intense in the 2-cm. circle in the center of the 5.5-cm. lighted circle and graded into dim light peripherally. Almost as soon as light struck the clams, they started crawling about randomly, the general direction of movement being outward away from the center of the light. In ten minutes half the clams had crawled out of the 2-cm. circle of bright light; in twenty minutes most of the clams had left and concentrated in the dimmer peripheral light; a few crawled to the outer dark confines of the dish. In the control dish at the end of the twenty-minute period, the plantigrades remained aggregated in the center of the dish; only a few had moved outward about 1.5 cm. from the original 2-cm. circle of concentration. Different size classes of plantigrades were about equally distributed throughout the zone of dispersion, indicating that all sizes tended to move away from the bright light at about the same rate.

The experiment was repeated with plantigrades ranging in length from 1.0 to 2.4 mm., twenty-five in each of the control and experimental dishes. In twenty minutes all the clams in the lighted dish but one, which remained closed, crawled out of the lighted 2-cm. circle, whereas those in the control remained clustered within the original 2-cm. circle. The degree of dispersion was not so marked in larger plantigrades as in the smaller ones. Hunter (1949) reports that young of *Hiatella* are also negatively phototactic; and Morton (1960) reports that *Lasaea rubra* becomes negatively phototactic at light intensities not much above those of twilight.

These results indicate that when out of sediment, plantigrades of *M. mercenaria* crawl away from light of moderate intensity into areas of dim light, and as they develop siphons and move deeper into the sediment they are less affected by bright light. Whether they respond by moving deeper into the sediment on bright days as at low tide in intertidal areas in nature is not known.

RESPONSE TO FLOW OF WATER. The fact that in some instances plantigrades opened when held in a dish devoid of sediment and started crawling when sea water was forcefully ejected over them from a bulb pipette suggested that they might exhibit a rheotactic response. We tested this during the summer of 1956 in North Carolina. Fifty-five plantigrades ranging in length from 1.0 to 2.4 mm. were placed in a flow trough eight feet long and four inches wide, about two feet downstream from the inflow. Sea water pumped directly from Bogue Sound (salinity 35 °/oo, temperature 28°C.) flowed at the rate of 2.5 cm./sec. in the trough and covered the clams to several times their length. Light from windows was arranged so that only twenty-five foot-candles struck the trough at right angles to the flow of the water. In a period of four hours only five clams had moved; these distances ranged from one to four centimeters from the original grouping; two clams crawled upstream, three moved downstream. At least under these conditions, then, no response to flow was detected. This might be anticipated, since in nature clams are generally faced by alternating ebbing and flooding currents rather than continuous unidirectional flows.

SUMMARY OF BEHAVIOR IN LABORATORY. When the pediveliger stage is reached, larvae alternate swimming and crawling, apparently seeking suitable setting surfaces. A negative response to moderate to bright light intensities, possibly combined with a positively geotactic response, may direct them downward onto the more dimly lit bottom. During the byssal plantigrade stage, after the swimming organ is lost, they affix most densely where sediment of fine sand and/or organic detritus lies thinly over solid surfaces. Photonegative and strong thigmotactic responses impel them to remain in contact with the bottom and, as the siphons develop, to burrow into the sediment. Thorson (1950) notes that the ability to postpone metamorphosis

and actively seek substrata is widely distributed among marine larval invertebrates.

Byssal Plantigrade Stages in the Field

FACTORS AFFECTING DISTRIBUTION AND DENSITY. In general observations on distribution of juveniles and adults of *M. mercenaria* in Great Bay and Little Egg Harbor during the period 1947–51, we often found clams concentrated most densely along the edges of channels and sloping portions of bars, among shells and living oysters, at the base of piling and similar baffles, in muddy sand in sod hollows offshore from eroding marsh banks, and in level bottoms characterized by slight irregularities.

At least the following factors may be hypothesized as instrumental in regulating density and temporal and spatial distribution of this bivalve on the bottom: (1) the rate of development of parental gonads, the time of spawning, and the duration of veliger stage, all affected at least in part by environmental temperatures; (2) the proximity and abundance of spawning parents and volume of spawn and the survival, transport, and degree of dispersion of the veliger stage by water masses from parents to setting sites; (3) the attraction of pediveligers to gradients of turbulence surrounding objects extending from the bottom which offer resistence to tidal flow; (4) the nature of the bottom and the intensity of light at the time of setting; (5) swimming by pediveligers and crawling by byssal plantigrades subsequent to initial settling to the bottom, and movement and concentration of byssal plantigrades affixed to organic fragments and fine grains of sand by swift tidal currents; (6) depredations by predators. The multiplicity of these factors undoubtedly accounts for the unpredictability of time and site of setting and subsequent density and distribution of this mollusk.

Information on behavior of pediveligers and byssal plantigrades in the laboratory and on the ecology of larvae in the field has been presented in earlier sections and sheds light on some of the points listed above. The following paragraphs will report field studies of post-set clams and provide additional information on their behavior, distribution, and density.

In the summer of 1949 between mid-July and the end of August, we made a careful study of the distribution and density of setting of *M. mercenaria* in Lower Little Egg Harbor. The largest veliger swarms were detected in late June and early July (Fig. 25); thereafter very little spawning was noticed. In deep water, sediment samples were collected with the Petersen grab. Ten full grabloads constituted a sample at any station. Samples were dumped onto screens suspended in a metal sluice set across our motor boat and then washed with a stream of sea water from a pump in the boat. In shallow water, samples were taken by hand and washed on hand screens. Mr. Pomeroy collected the samples and provided me with a record of his field observations.

The lengths of plantigrades collected ranged from one to several millimeters. Sets occurred widely though irregularly throughout the bay. Their distribution did not coincide entirely with that of adults, for there were some bottoms with dense populations of young adults in which no plantigrades were found. The maximum concentration, 125 byssal plantigrades per square meter, was located on a shell bar in Thompson Creek (Fig. 14); the next highest, 54 per square meter, was taken in the central region of the bay. Dissoconchs were measured in anticipation of recording growth rate. Size range was extreme, however, and no measurements of any day's collection fell into a normal curve of distribution; the curves assumed bi- or trimodal or irregular form. This irregularity probably reflected the occurrence in time of a series of sets on the same bottom as well as wide limits of size variation of different age classes (as demonstrated in laboratory growth studies). Thus it was not possible to obtain reliable progressive size increments from the data.

The dense set in Thompson Creek provided information on behavior and mortality rate of byssal plantigrades. The set was discovered initially on July 12 on an artificial shell bar in the mouth of a tidal creek tributary to Thompson Creek and near the "Cynthia." A census on July 13 showed that dissoconchs ranged in length from one to four mm. The maximum population density, about 125 clams per square meter, was located on the shallowest portion of the bar, which also held the heaviest covering of oyster shells; density decreased down the sides of the bar, and plantigrades were quite infrequent on the unshelled mud bottom around the bar.

Mr. Pomeroy noticed that plantigrades were found principally in single upturned oyster shells containing mud (mixture of organic detritus and fine sand and silt). This mixture was flushed gently from the shells into a screen with a bulb pipette; flushing uncovered the plantigrades, which remained affixed to the shells, even to sites covered by more than a

centimenter of mud. Plantigrades were not visible on the surface of the mud or on clean shell, apparently having selected surfaces covered by sediment. Mud under and around the shells contained but a fraction of the number found in the shells. These observations confirm those made in the laboratory and suggest that in the field, byssal plantigrades seek out available solid surfaces on which to anchor, surfaces which are coated by sufficient loose material to permit them to burrow as soon as their morphology permits. These ecologic requirements may be satisfied by the presence around hard surfaces of a variety of mixtures of fine sedimentary grains. Coarse gravel or shell or organic mud alone do not seem to constitute as desirable setting sites. In firm sandy or sandy mud bottoms devoid of hard substrates, as in Lower Little Egg Harbor, plantigrades make multiple attachment to several grains of sand at a time. A dense set of *M. mercenaria* on an intertidal bar in Shark River channel, some 40 miles northward along the New Jersey coast, made possible observation of the position of byssal plantigrades about 3.5 mm. long in clean fine sand. The activity of dissoconchs was observed as incoming sea water rose over the bar to a depth of a half-inch. Plantigrades were buried to about one-half their height, and they clung to surrounding sand grains by means of thin, branching byssi, as described by Belding (1912) for young clams in Massachusetts, their short definitive siphons and siphonal sleeves fully extended as they actively pumped water. The role of clean, unstable sand and larger particles in setting, concentration, and survival of plantigrades in channels and inlets of swift tidal flow has already been discussed (Carriker, 1959; Turner and George, 1955); in general, byssal plantigrades are well adapted for life in these shifting sediments.

After several weeks we noted that plantigrades on the Thompson Creek shell bar were no longer as closely associated with shell surfaces. Examination of upturned oyster shells, as in the preceding survey, revealed the presence of only a few plantigrades, whereas screening of sediment around the shells revealed many more. Two explanations suggest themselves: either dissoconchs, on reaching a certain size or age, crawl from the shell into the surrounding sediment, or those in cupped shells are largely removed by predators, whereas those in the sediment around the shells escape enemies in greater measure. Possibly both factors operate, their degree of importance correlated with the type of bottom materials and the number and kinds of predators present.

By July 30, some two weeks after the initial survey of the artificial shell bar, mortality of plantigrades on the bar approximated 50 per cent. Other areas in the bay where successive sampling was carried out demonstrated a similar trend in mortality.

Since the plantigrade population on the shell bar was already some weeks past the setting time when first discovered, its density, because of inroads of predators, undoubtedly represented but a fraction of the original one. An effort was made to determine the original setting concentration on the basis of living dissoconchs and fresh shell fragments. Bottom material on the bar was screened through nested screens of 1 and 0.5 mm. pore size. Living plantigrades and fresh empty valves and valve fragments were removed and recorded. After several weeks of this study, the initial population density (that at July 13) was compared with those estimated from subsequent analyses. The conclusion was that even if every single valve and fragment were counted as an individual, the figure seldom exceeded 50 per cent of the density of the population at discovery. Furthermore, it was soon apparent that the quantity of empty valves and fragments recovered varied widely with nature and location of bottom, since delicate material of this kind is subject to removal by tidal currents and to rapid destruction by abrasion induced by waves and currents. Thus collection and sorting of dead shell material gave some information on the nature of predators, but proved unreliable for determining the density of the clam population at time of setting.

As a sequel to study of the distribution of byssal plantigrades in Thompson Creek, it is worth reporting a detailed study made by the writer in 1948 of the distribution of a dense population of hard clams in a system of old baffles on Edge Cove Bar. Local clammers said that they had not harvested Edge Cove, and our preliminary sampling disclosed a population represented by several year classes, ranging in length from a few millimeters to 8.4 centimeters. Although the population on the bar was relatively undisturbed by man, it was subject to predation by at least the following animals common to the area: *Busycon canaliculatum, Urosalpinx cinerea, Callinectes sapidus,* and xanthid mud crabs.

Edge Cove Bar runs eastward from the shore of Edge Cove (Fig. 34) about 200 feet; it is 40 feet wide, about a foot above the level of

the cove and it is composed of a mixture of coarse and fine sand, shell, and mud overlying the firm marsh mud with its entanglement of partly decayed *Spartina* roots. Many years ago an extensive system of vertical and horizontal piling was installed for a dock and buildings, and the bottom was hardened by addition of sand and shell (T. C. Nelson, personal communication). At the time of this study many of the old timbers were still on the bar and afforded several combinations of baffles, boards, and posts ranging in height from the level of the bar to five feet with diameter up to eight inches. Most conspicuous were two rows of piling running the full length of the bar (Fig. 37); short rows extended at right angles from

FIG. 37. Photograph of major rows of baffles on Edge Cove Bar, Little Egg Harbor, N. J., at mean tide level (compare with Figure 15: 1948).

these, and single piles and clusters of piling were scattered over the bar. The bar sloped gradually from the beach to a depth at mean low water of approximately 30 inches at the outer end; during extreme low tides coinciding with strong westerly winds, the inner third of the bar was exposed. Tidal currents moved across the bar into tidal creeks at right angles to the principal rows of baffles. Current velocity, as measured through the cycle of an average tide, attained a maximum velocity of 10 cm./sec. over the outer end of the bar, and about 8 cm./sec. midway out the bar. Rainfall, tidal amplitude, salinity, and water temperature for the Edge Cove area are summarized in Figure 15.

A census of the bar was made in the following way. During periods of low, calm water, sediment on the bar was first scraped from quadrats 1 × 3 feet at a time and washed on a screen of 1-mm. pore size. This operation collected small clams. The remainder of the quadrat was then searched to a depth of some six inches for larger individuals. Quadrats were chosen to include the range of types of bottom and baffles present. A total of 372 square feet of bottom was examined, and 1,178 clams were collected, distributed in the following size classes:

Length Range, mm.	*Number*
5–24	132
25–37	74
38–50	223
51–63	432
64–75	235
76–84	82

These figures suggest that setting of *M. mercenaria* over the previous eight or so years (age of the oldest clams approximated from number of annual growth rings) occurred rather consistently, though they cannot give much information on relative density at time of setting, since the variables of setting intensity and predation pressure are not necessarily correlated.

A census of the bar provided an average density of about three clams per square foot of bottom—a misleading figure, since density varied from 0 to 50 clams per square foot, dependent on presence or absence of baffles, depth of water, and nature and slope of the bottom. Maximum concentrations occurred in an area 100 to 200 feet from the shore in a minimum depth of water at mean low tide of about one foot, and always in sediment around baffles of all types: round piling, slats, stones, sunken logs, and shell. The highest density encountered—150 clams per three square feet—was found at a distance of 184 feet from the shore in two feet of water (mean low tide) around a post seven inches in diameter and three feet high, surrounded by a cluster of shorter, smaller stakes, stones, and shell in a mixture of mud and coarse sand. In comparable quadrats ten feet away but lacking shells and piling, no more than four clams were present. The type of piling, whether slats or round, did not appear to affect concentration, though more clams were found around higher baffles. Bottom littered with bivalve shells always contained more clams than similar bottom without shell, a phenomenon also observed on the Thompson Creek shell bar. Sand or a mixture of mud and sand or pliable mud generally contained more clams than comparable bottom consisting of hard marsh sod. Clams were also more dense where the nature of the bottom changed abruptly from hard mud to sand than in either the mud or sand, and more numerous along the marginal slopes than on the plateau of the bar.

The following points may be induced from the study of distribution of *M. mercenaria* on Edge Cove Bar: (1) Objects in the water, acting as baffles to currents, create current velocity and turbulence differentials which may stimulate pediveligers to set around the baffles in greater abundance than on bottom without such baffles. Since, however, the location of adult clams does not necessarily delimit the setting area nor their number indicate setting intensity, the survey provides only circumstantial support of this point. (2) Behavior of byssal plantigrades in the laboratory and on the Thompson Creek shell bar suggests that they tend to move into shaded microhabitats filled with shallow sediment, such as cupped shells, crevices in piling, depressions among sessile occupants of hard surfaces, and boundaries between hard surfaces and loose sediment. Thus they may crawl or swim into the vicinity of such baffles secondarily after settling to the bottom, in addition to those which set originally about the baffles in response to gradients in the turbulence of the water. Baggerman (1953) has shown that young cockles (*Cardium edule*) are transported by water masses long after metamorphosis and until they reach a length of 1.8 to 2 mm. (3) Baffle areas provide microhabitats which provide pediveligers and byssal plantigrades some protection from predators. Consequently, even if setting were uniform over the whole general area, survival would be greater in the immediate vicinity of the baffles. Probably all of these factors are functioning collectively, though in varying degrees in different environments. For example, attraction of pediveligers to gradients of turbulence may be maximal in areas of swift tidal currents, while in regions of more sluggish circulation secondary movement of young clams to the protective baffle site may play a more important role in concentration there. And in areas of unstable sediments and swift tidal currents rate of predation is relatively low as compared with that in more quiet areas (Carriker 1959).

Belding (1912) noticed that sets of *M. mercenaria* often occurred in eddies or on the sides of a swift current. Orton (1937), in a study of the intensity and distribution of spatfalls of *Cardium edule* in Morecambe Bay, notes that these bivalves "become concentrated and precipitate out of the plankton in localities where slack and eddy waters occur." The possible role of the action of water motion in concentrating setting pediveligers is discussed briefly by Turner (1953), and as he points out, since we do not yet know the response of pediveligers to gradients of turbulence under controlled conditions in the laboratory, we are not in a position to do more than hypothesize on the role of turbulence in vertical distribution and setting of these larvae.

EXPERIMENTAL STUDY OF SETTING IN THE FIELD. Because byssal plantigrades soon mingle with surface grains of sediment after metamorphosis, it is difficult to study rate and intensity of setting, growth rate, and survival in the field. During the five summers in Great Bay and Little Egg Harbor we developed a setting indicator, based on Thorson's (1946) "bottle-collector" for gathering newly set sessile invertebrates, by means of which it was possible to make a beginning in such studies.

The clam-setting indicator was evolved to take advantage of those characteristics in the setting behavior of pediveligers which would stimulate them to affix in the indicators: (a) possible attraction to gradients of turbulence about objects in water, (b) preference during daylight for microhabitats in subdued light, and (c) preference for contact with shallow layers of fine sediment over hard surfaces. The device protects plantigrades from outside predators without deleteriously impeding flow of tidal water, food, and oxygen over them, so that they feed and grow normally in it. The indicator may be removed from the water with a minimum of disturbance and the plantigrades within it studied alive in the laboratory and returned in the indicator to the water. The device further makes possible relatively easy detection and manipulation of young plantigrades in a minimum of fine sediment.

Setting indicators were constructed from the upper third of narrow-mouthed, one-gallon glass bottles six inches in diameter across the inverted large end (Figs. 38, 39). A one-hole rubber stopper with a five-inch paraffined, galvanized, large-headed nail in it was inserted in the neck of each bottle top. A piece of wooden doweling eight inches long and seven-eighths inch in diameter fit snugly into the neck of the bottle top, and plastic screening (pore size approximately 1 mm.) was wrapped over the doweling and bottle top and secured at the neck with plastic twine. Plastic screen and twine were unaffected by exposure to sea water over periods of several months.

Indicators were supported on a frame which made it possible to place them anywhere in the shallower portions of Lower Little Egg Harbor. The frame consisted of (a) a long, stout, cedar guide pole about three or four inches in diameter

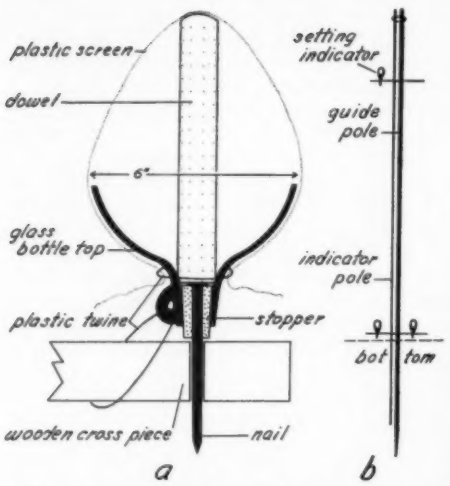


FIG. 38. a. Drawing of hard-clam setting indicator. b. Drawing of support for hard-clam setting indicator.

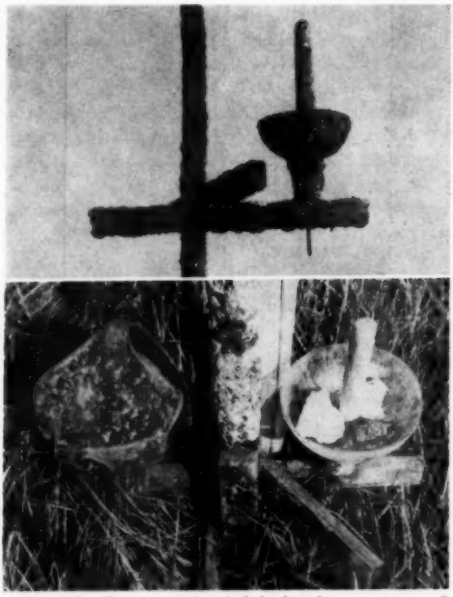


FIG. 39. Photograph of (a) (top) an uncovered setting indicator in position on the top rung of an indicator support after exposure to fouling organisms; (b) (bottom) two indicators on bottom rung of support; the one at left covered by screen, the one at right uncovered and containing a variety of substrates used in early experiments and later found unnecessary.

at the slender end which was driven stout end first into the bottom to a sufficient depth (some three feet in soft bottom) to provide secure anchorage for the indicators and temporary moorage for a small boat and be visible above water at high tide; and (b) a long, light, slender indicator pole, likewise pointed at the stout end, with two sets of cross pieces nailed to it, one set located near the lower end of the pole and intended to rest on the bottom, and the second set so placed as to remain at least a foot or two below the surface of the water at low tide (Fig. 38b). Indicators were slipped into holes drilled on the outer ends of the cross pieces and held in position by plastic twine looped around the cross pieces and handles of the bottle tops. In operation the indicator pole was guided down the guide pole to, and pressed a short distance into, the bottom. The two poles were then fastened together at the top with heavy twine. When indicators were inspected, the indicator pole was brought carefully into the boat in a vertical position, and indicators were untied and lifted from their supporting cross pieces.

Bottle tops were selected for indicators because (1) glass provides a hard, easily cleaned, internal surface, (2) the tops can be lifted from the bay without disturbing the contents unduly, and (3) plantigrades and sediment may readily be dislodged (with a thin-edged rubber spatula or test-tube brush) and rinsed (with a large bulb pipette) into beakers for analysis in the laboratory. Some of the hard plastic bottles now on the market may serve as well as the glass and contribute the advantage of unbreakability. After screening and counting, sediment and plantigrades may be returned to bottle tops and replaced in the bay, or plantigrades may be examined directly in the bottle top under a microscope after sediment has settled and water has cleared. Plantigrades affix to glass within the bottle top principally along the advancing edge of accumulating sediment and may be located there after most of the sea water in the bottle top has been siphoned out.

The cone of plastic screen over the bottle top, which excludes predators, reduces light intensity and creates a differential in rate of water flow over the bottle top, was cleaned of fouling organisms by brushing. Some screens which had been painted with commercial copper paint to reduce fouling remained free of most fouling organisms for at least two months of submergence but may have reduced the quantity of clams setting in the indicators. Bottle tops

Table XIII

Schedule of placement and examination of clam setting indicators and density of set per bottle top at Station 4 in Lower Little Egg Harbor.

Location of top and type of screen	Dates of Placement and Examination of Tops							
	July							August
	3	7	11	17	21	27	31	28
(1) Bottom, unpainted.....	Set	0	0	0	0	—	0	discontinued
(2) Bottom, unpainted.....	—	—	—	—	—	Set	1	202
(3) Bottom, Cu-painted.....	Set	0	0	1	0	—	0	125
(4) Surface, unpainted.....	—	—	—	—	—	Set	1	140

were examined in the field during periods of relatively quiet water, since rough water caused the boat to pitch and roll, spilling the contents of the indicators.

A field test of the fully developed clam-setting indicator was first made during the summer of 1951 at Station 4 in Lower Little Egg Harbor. Four indicators were placed on one indicator pole as follows: three (one with a copper-painted screen and two with unpainted screens) near the bottom with the rim of each bottle top about six inches above the surface of the bottom, and one (with unpainted screen) four and a half feet above the bottom in water seven feet deep at mean low water. The schedule of setting and examination of the indicators is presented in Table XIII.

The following procedure was employed in examining effectiveness of bottle tops as setting indicators. Sediment and occupants were brushed and rinsed into glass dishes and transported to the laboratory. Plantigrades remained alive for at least four days in these dishes, even though they were thoroughly mixed in sediment at the start and had to crawl to the surface through some two centemeters of organic mud. In the laboratory sediment was flushed in small quantities through two nested stainless steel screens, No. 60 (pore size 260–340 μ) and No. 100 (165–210 μ). Residue from each screen was then rinsed into a large, deeply concave dish (bottom portion of a Kjeldahl flask), where it was swirled circularly by the force of water ejected from a large bulb pipette. Plantigrades, being relatively heavy, remained on the bottom, which permitted floating materials to be drawn off. Mechanical vibration caused by the handling of containers tended to keep dissoconchs closed; however, as a precaution against the possibility that pediveligers might start swimming and be drawn off with floating residues, brackish water instead of sea water was employed in screening and swirling. Sediment and animals remaining after swirling and cleaning were rinsed into a shallow watch crystal about four inches in diameter and concentrated in the center of the dish by further swirling. Materials held on the No. 100 screen were examined under a magnification of 25X, and those on the No. 60 screen under 15X magnification. A dissecting needle bent slightly and filed flat at the tip on opposing sides facilitated sorting and movement of sediment under the microscope. Most dissoconchs, possessing a density slightly less than that of sand, were concentrated at the outer edge of swirled materials. Where excessively large quantities of sediment collected in bottle tops, aliquot fractions were examined.

The relation of the density of set obtained in indicators and the density and temporal distribution of larvae in the bay may be seen by comparison of Table XIII and Figure 27. Although dense swarms of larvae appeared during the period July 10–13, no late stages of these larvae were recovered in plankton samples, and only negligible setting took place: one pediveliger 200 μ in length was taken in bottle top (3) on July 17. Since clean bottle tops (1) and (3) had been placed in the bay only six days earlier, there is a possibility that insufficient sediment had accumulated in them by the time pediveligers came by. This suggests that bottle tops be inoculated with a small quantity of fine-screened sediment before placement in the water. In spite of the fact that large swarms of clam veligers present in the water on July 23–25 remained in the water in small numbers through the late umbone stages, setting in bottle tops was still negligible (see July 31 examination, Table XIII). Two explanations are suggested: either setting occurred in other parts of the bay, or there was insufficient sediment in the bottle tops to attract those pediveligers which passed by.

The principal set of the season in bottle tops was found on August 28 (Table XIII) after the indicators had been unattended for four weeks. At this time the indicators on the bottom were filled to within three-fourths inch and the one near the surface to within two inches of the rim with fine sediment which entered in suspension and settled as the flow of water decelerated in the bowl of the indicator. The maximum density was found in the bottom indicator with unpainted screen (202 plantigrades), intermediate density in the indicator near the surface (140), an minimum density in the bottom indicator with the copper-painted screen (125). These data suggest that pediveligers may be distributed throughout the vertical column of water and that there is setting at all depths, with no clear tendency for greater setting at any depth. Furthermore, copper paint may tend to repel pediveligers.

The area of the open end of each bottle top was 28 square inches, or 0.2 square foot. Thus (though the optimal setting surface within each bottle top, the shallow shore around the accumulating pool of sediment, was many times less than the area of the opening of the top), the density of set ranged from 625 to 1,010 plantigrades per square foot of bottle top. Since concentrations of this magnitude were never found in natural bottoms in the area, it may be inferred that indicators functioned in at least two ways in compounding such densities: (a) by excluding macroscopic predators and to a large degree eliminating predatory loss of plantigrades, and (b) by concentrating pediveligers which were stimulated to enter and to remain, attracted by conditions in the indicator. It is doubtful that pediveligers were trapped in the sense that they could not leave after entering the device, since they should have been able to swim out as readily as in, and after loss of velum to creep out through the screen, though once valves exceeded dimension of pores in the screen (approximately 1 sq. mm.), plantigrades were caged.

The size of plantigrades taken in indicators fell in two major length classes: (a) 3 individuals 900–1,400 μ, which on the basis of rate of growth of plantigrades in our laboratory, came from the spawning of July 30 (Fig. 27); and 464 individuals 208–340 μ, which came from the spawnings of August 9 and 18. Late-stage umboned veliger larvae were recovered from the water after all these spawnings, and in more than usual abundance after the August spawnings. Thus intensity of set in the bottle tops was roughly proportional to abundance of late-stage veligers in the plankton.

The distribution of plantigrades in these indicators and in earlier, less effective models in which a variety of setting surface was provided demonstrated that the preponderance of setting occurred on hard surfaces overlaid by shallow layers of sediment. This confirmed observations made in the laboratory and suggested elimination of "cultch" of any sort from the bottle tops (Fig. 39b, right), thus greatly simplifying the construction and use of indicators.

Bottle tops were successful in accumulating other invertebrates. Barnacles, serpulid polychaetes, xanthid mud crabs, and the bivalves *Teredo, Mulinia, Modiolus, Tellina,* and *Tagellus* were commonest. Hardiest of the bivalves were *M. mercenaria,* which remained alive longest in dishes in which the content of indicators in some instances was stored for several days. As many as four or five young mud crabs up to six mm. in width were found in all indicators on August 28. These entered as plankters and grew, and then could not escape. The age they commence preying on young bivalves, or whether they destroyed any plantigrades in the indicators, is unknown. The large concentration of dissoconchs in bottle tops would suggest that little if any predation did occur. Studies showing at what size or age different species of crabs begin attacking young hard clams would be helpful in establishing a time interval between placement and examination of indicators in order to eliminate predation by young crabs.

Clam-setting indicators were employed in Home Pond (Carriker, 1959), but pediveligers were not present in sufficient quantities at the time to produce more than an occasional plantigrade in the indicators.

IV ENEMIES

Hard clams of all ages are hardy animals tolerant of a wide range of physical environmental conditions. Of all the stages in the life cycle, veligers are considered the most sensitive, and yet these are successful within a broad range of many factors. Davis (1958) reared shelled veligers to metamorphosis in constant salinities ranging from 17.5 to 35 °/oo in Connecticut; in North Carolina the maximum salinity of the water (which did not drop be-

low 31 °/oo) in our cultures was 36 °/oo. Loosanoff et al. (1951) and Loosanoff (1959) in Connecticut grew larvae to setting at constant temperatures ranging from 18° to 30°C.; and in New Jersey the temperature of the water in our larval cultures, fluctuating with air temperatures, ranged from 13° to 30°C.; in North Carolina maximum temperatures reached 31°C. Since post-set stages of hard clams in the field occur in sediment types ranging from clean sand to organic detritus, in bottoms varying from those beneath sluggish wind-driven circulation to inlets with extremely swift tidal currents, and in various degrees of exposure in intertidal zones, it may be concluded that pediveligers and byssal plantigrades likewise withstand at least this spectrum of conditions.

However, *M. mercenaria*, particularly in the early stages, is attacked by a surprisingly long list of organisms. In this section we report our observations, and those of other investigators, of enemies of young stages of this bivalve.

PLANKTON FEEDERS. Thorson (1946), from an exhaustive study of the planktonic larvae of Danish bottom marine invertebrates, concludes that the striking loss of these animals before and during setting is due only in small measure to lack of fertilization of eggs, starvation, extremes of environmental factors, or unsuitable substrates, since probably all are able to postpone metamorphosis for some days or weeks, providing time for selection of suitable setting surfaces. Major losses, he believes, are probably due to a large number of planktonic, pelagic, and benthic animals which feed in part or entirely on planktonic larvae (Thorson, 1950).

We did not have opportunity to examine the stomachs of marine animals associated with *M. mercenaria* in Little Egg Harbor. But on the basis of the observations of others as summarized by Thorson (1946), and animals present in Little Egg Harbor, it is reasonable to suspect at least the following as predators of one or more of the planktonic and newly set stages of *M. mercenaria* in Little Egg Harbor: *Noctiluca, Aurelia, Diadumene, Mnemiopsis,* decapod zoea, *Balanus, Chthamalus, Crassostrea, Mercenaria, Mytilus, Modiolus,* and *Brevoortia.* In this connection Belding (1912) notes that "all manner of fish, crustacean and molluscan life feed on the larvae, even the mother quahaug sucking down her own offspring." This array of predators could well produce the high mortality of hard-clam veligers we observed in Little Egg Harbor. Furthermore, low concentration of veligers near the bottom may be explained in part, perhaps, by depletion of larvae by filter-feeding predators there, though that that is not the whole explanation is indicated by the vertical movements of the larvae. As Thorson points out, the volume of water which these bottom-dwelling predatory forms filter per day must be a sizeable fraction of the total volume of water present. Rice and Smith (1958) have determined that for *M. mercenaria* alone in suspensions of diatoms the average filtering rates range from 3 to 6 liters per hour, and in suspension of green algae, as high as 2 liters per hour.

FUNGI. *Sirolpidium* sp., first described by Davies et al. (1954) in laboratory-reared veligers and identified by us in our larval cultures on Gardiners Island, on occasion reached epidemic proportions. As described by Davis, the majority of the infected larvae stopped growing and died soon after infection. We did not encounter the fungus in laboratory cultures in North Carolina.

PROTOZOA. Loosanoff (1959) reports that the large heterotrich ciliate *Condylostoma* sp. ingests hard-clam veligers when present with them in his laboratory cultures, a single ciliate consuming as many as six larvae. We did not find this organism in our cultures.

DECAPOD CRUSTACEA. As soon as the high mortality of byssal plantigrades was noted on the Thompson Creek shell bar, we made a search for their major predators. Suspected predators were isolated, each species with a group of plantigrades, in small screen cages suspended in the water from the "Cynthia."

Of the xanthid mud crabs—*Eurypanopeus, Neopanope, Panopeus,* and *Rhithropanopeus*—in the area, *Rhithropanopeus* was most common in the slightly brackish waters of the creeks and coves along the western shores of Little Egg Harbor, and was therefore used in the experimental cages. In some bottoms the density of mud crabs exceeded that of byssal plantigrades. Crabs were divided into two size groups, and each was given hard clams ranging in length from two to about nine mm. in length. Crabs having a carapace width of 1 cm. crushed dissoconchs less than 3 mm. long; crabs 2 cm. wide crushed dissoconchs 6 mm. long. These decapods, and especially the larger size, were voracious: one in a cage with fifteen clams consumed thirteen in three days. Flesh was obtained by completely crushing the valves with the chelipeds, leaving a residue of clean irregular shell fragments.

Depredation of juvenile *M. mercenaria* by Xanthidae, *Callinectes sapidus*, and *Carcinides maenas* has been reported by several authors (reviewed by Carriker, 1959). Although predation of byssal plantigrades by young blue and green crabs has not been recorded, it is likely that much does occur, and the possibility merits study.

MEROSTOMATA. No large numbers of *Limulus polyphemus* were present in any of our study areas, but T. C. Nelson (personal communication) reported that on the intertidal flats of Cape May, New Jersey, where the horseshoe crab was abundant, densities of newly set *Mya* running to a maximum of 9,000 per square foot were completely destroyed by decapod crabs and horseshoe crabs in a few weeks. Shuster (1950) writes that small specimens of *Limulus* feed on minute organisms that live in the surface sediment, and that as they grow they feed on larger prey; the diet of adults consists chiefly of marine worms and several species of bivalves. This animal, then, may be considered a serious potential predator of at least younger stages of *M. mercenaria*.

MURICID GASTROPODA. Examination of empty hard-clam byssal plantigrade valves collected in the field in Little Egg Harbor disclosed the presence of many small relatively straight-walled holes in the shells. The only boring gastropods known to exist in the area were *Urosalpinx cinerea* and *Eupleura caudata*. Young snails isolated in experimental cages with byssal plantigrades bored holes similar to those encountered in empty valves in the field. Location of the holes in valves varied widely, and not infrequently snails penetrated between the valves, leaving a hardly visible crescentic nick on each valve.

Subsequent laboratory studies (Carriker, 1957) in North Carolina demonstrated clearly the voracity of newly hatched *Urosalpinx cinerea* and the degree to which young disso-conchs of *M. mercenaria* are drilled and consumed. These young conchs exhibit a pronounced positive chemotactic response to the external metabolities of young clams, and readily locate them. A snail 1.5 mm. in height perforated a clam 0.6 mm. in length in 45 minutes and rasped out the flesh in an equal time. Predation rates varied from one to nineteen young clams drilled and consumed per conch per day. (However, it should be emphasized that in the field other species of bivalve prey would constitute buffer-food species which would tend to reduce the rate of predation of clams.) Young *Urosalpinx* can crawl through superficial layers of sediment, thus easily reaching those young clams which have burrowed shallowly after formation of definitive siphons. Although veliger and protoconch stages in these snails are passed in an egg case, young conchs may be widely distributed by crawling onto debris and floating on it in tidal circulation (Carriker, 1957).

NATICID GASTROPODA. Two common species of this group of boring snails—*Polinices duplicatus* and *Polinices heros*—are encountered in habitats in association with hard-clam populations. In Little Egg Harbor the former were abundant on the bay side of the barrier beaches. The perforation drilled by these species differs characteristically from that by muricids listed above in being distinctly and broadly bevelled. Hanks (1960) reports that recently metamorphosed *Polinices* are able to bore into small bivalve prey. H. N. Gibbs (personal communication) observed moon snails 2 mm. in diameter, and we saw specimens 4 mm. in diameter, drilling young bivalves.

The high rate of predation of burrowing bivalves by adult naticids has been reported several times (see review by Sawyer, 1950; Carriker, 1951; Ansell, 1960; Hanks, 1960); buried prey find no protection in sediment, since moon snails themselves are such facile burrowers. Only one published report is available on predation by young moon snails. Sawyer (1950), utilizing *P. duplicatus* ranging in diameter from 9 to 12 mm. in laboratory experiments in Massachusetts, learned that at a water temperature of 21°C. snails drilled and consumed 0.67 individual of *Mya arenaria* per day. Increase in weight of the snails during a thirty-day feeding period approximated one-twentieth of the weight of the clams consumed during that time.

A high rate of predation combined with a pelagic larval stage and an unusual method for pelagic distribution of young conchs (Shuster, 1951) make young *P. duplicatus* formidable enemies of young bivalves. During the first week in August, 1951, Shuster observed young snails 4.0 to 9.4 mm. in diameter concentrated on sand bars 400 yards offshore on the New Jersey Cape May intertidal flats. Three weeks later hundreds of individauls of the same population, now ranging in diameter from 6.4 to 16.5 mm., were seen with foot fully expanded floating and drifting across the tidal flats on the shallow incoming tide. As water flooded the flats, snails lifted the propodium, curled the sides of the foot downward and inward, and rolled over on their sides. They floated readily

in this position with a large sheet of mucus passing from the extended foot.

OTHER GASTROPODA. The capacity of adult *Busycon carica* and *B. canaliculatum*, both common associates of *M. mercenaria*, to penetrate hard clams by mechanical chipping has been described, and a preliminary estimate of the rate of predation in New Jersey has been recorded (Carriker, 1951). Probably other species of whelks penetrate tightly closing bivalves in a similar manner, though this should be verified. The size at which young whelks first attack young hard clams has not been determined, but it is probably soon after emergence of the protoconch from the egg case.

Young of several species of browsing snails—like *Nassarius obsoletus*, which occur in such abundance in hard-clam setting areas—though not predators (Jenner 1956) may inadvertently consume early byssal plantigrades with detritus and microscopic organisms as they rasp over the surface sediments with the radula. Preliminary observations by us, and independently by Turner and George (1955), indicate that the mud snail does not consume clams 0.5 mm. or more in length. However, the study of accidental consumption of newly set byssal plantigrades by browsing mud snails may show a different picture and should be studied.

ECHINODERMATA. Sea stars like *Asterias forbesi* and *A. vulgaris* may occur in conspicuous abundance (Burkenroad 1957) in areas where *M. mercenaria* is likewise found, and the damage which these adult asteroids inflict on macroscopic epifaunal bivalves is well known. Whether recently metamorphosed sea stars attack byssal plantigrades of hard clams before they burrow beneath the surface of the sediment has not been investigated. It is likely that they do.

TUNICATA. On Gardiners Island larvae of *Molgula manhattensis* entered in running sea water and settled in trays in the laboratory. As the tunicates increased in size the valves of hard-clam byssal plantigrades became fastened to their tunics. In one instance we found six ascidians with some 150 plantigrades, of which about 5 per cent were dead, stuck fast to and partly overgrown by the tunic. It would appear that hard clams were attracted to the hard surface provided by the tunicates, attached by means of the byssus, and then became embedded in the tunic as it enlarged. Whether deaths were brought about by this is not known. Sea squirts of this species settling in trays of young hard clams in the field collected plantigrades about them in a similar way. Ryder (1889) reported the attachment by the byssus of large concentrations of *Mya arenaria* to the tunics of *M. manhattensis* in the field and did not notice adhesion of the valves to the tunic. Polychaete larvae (*Polydora* sp.) also entered our laboratory trays, and as they grew constructed tubes of detritus which did not, however, affect the young clams.

OSTEICHTHYES. Bigelow and Schroeder (1953) report that the puffer *Sphaeroides maculatus* (an inshore, shallow-water summer fish found along the Atlantic coast in abundance) includes small mollusks in its diet. Young fry 7 to 10 mm. in length collected near Woods Hole contained "molluscan larvae" (presumably early plantigrades) in the stomach. T. C. Nelson (personal communication) has observed puffers in shallow water in the field, inflated with water and upended over the bottom, washing young clams out of the sediment with streams of water jetted from the mouth. In Little Egg Harbor we caged a small puffer with young hard clams in an attempt to study this behavior, but the fish did not feed, perhaps because of confinement in the cage. Further observations are indicated.

ANSERIFORMES. Some ducks and geese are reported to consume small marine bivalves (Martin and Uhler, 1939). These birds which congregated on ponds on Gardiners Island during the winter thoroughly worked over the shallow bottom with feet and bills, leaving it pockmarked by craterlike depressions two to six inches deep and up to a foot in diameter (Carriker, 1959). Determining the quantity of young hard clams consumed in this way was not possible but it is probable that many were sieved from sediment by the birds.

V DISCUSSION AND CONCLUSIONS

Mercenaria mercenaria is a typically American species found principally along the eastern seaboard of North America. Its habitat is the more saline coastal embayments and estuaries; only occasionally does it appear to survive in the open ocean, and then it is found in the close vicinity of inlets.

Possessing a planktonic larval stage, the hard clam is confronted simultaneously with the advantage of dispersal of its young to widely

available suitable substrata in embayments and estuaries and with the potential danger of transport out to sea to unsuitable habitats. To what degree the latter occurs is known only in a general way and is undoubtedly dependent upon the specific circulation patterns of each coastal body of water in which the clam resides.

This paper presents information on the role played by functional morphology, behavior, and autecology in fitting early stages of *M. mercenaria* to remain and survive within the complex gradients characteristic of coastal habitats.

In the laboratory trochophores and earliest straight-hinged veligers concentrate noticeably at the surface of culture jars, and presumably the same occurs in nature, though to date the opportunity to observe this has not occurred. As soon as the straight-hinged veliger acquires valves, its specific gravity considerably exceeds that of sea water, so that the larva sinks bottomward as the velum is withdrawn. So far as is known, velar activity persists throughout the planktonic life of the veliger, the velum increasing in size and effectiveness as the veliger increases in size and weight and approaches the pediveliger stage. Though the larva cannot prevent horizontal dispersal, it does actively seek and maintain a vertical position in the water off the bottom and away from bottom-dwelling predators. The veliger constantly faces extinction by pelagic and nektonic filter-feeding animals in the water, but enemies on the bottom are generally more abundant. Even during hours of darkness, when veligers assume a broader vertical distribution and some are taken near the bottom, the zone of maximum larval density occurs in mid strata. Whether the scarcity of veligers near the bottom reflects predation by bottom-dwelling organisms or whether (more likely) the larvae have evolved an as yet undescribed behavioral response that tends to keep them swimming off the bottom—or both—is not known. The significance in terms of survival value of such a response does not require amplification. Further, it is conceivable that some of the diurnal differences in vertical distribtuion of hard-clam veligers may reflect those of food organisms, veligers migrating to zones of maximum concentration of food organisms as they feed. These points have not been demonstrated and should be investigated.

The tendency of adult hard clams to spawn during neap tides and close to ebb slack water in Little Egg Harbor favors retention of the larvae in the bay. During neap tidal periods minimum exchange of water in the bay takes place with that in the ocean, thereby reducing loss of larvae to sea by exchange. Larvae resulting from spawnings at ebb slack tide are transported up the bay during the subsequent flooding tide rather than down the bay toward the inlet; this favors especially those larvae spawned within the horizontal length of a tidal excursion from the inlet.

A short veliger stage, a minimum of eight days in both the laboratory and the field, also confers survival value, enhancing the chance for metamorphosis to occur within the bay before exchange and flushing transport veligers into the ocean. Veligers which appear in the bay coincident with periods of minimal precipitation are further favored in that they face less danger of transport to sea than those confronted with high flushing rates resulting from heavy precipitation. The degree, if any, to which precipitation influences the time of spawning has not been established.

Nor has it yet been determined whether veligers of hard clams, like those of oysters, exhibit differential vertical distribution in the course of veliger development which results in transport of older stages in the long axis of the estuary by nontidal water movements. Distribution of adult hard clams up the Great Bay estuary does suggest that some such mechanism may operate there.

It is probable, though this requires verification, that *M. mercenaria* upon assumption of the late pediveliger stage becomes photonegative, geopositive, and attracted to substrates.

Once the post-pelagic stage has established itself in the bottom, the hard clam moves only short horizontal distances (Chestnut, 1952). For the remainder of its life it is bounded by a narrow substratal horizon and the water mass of a tidal excursion. But early bottom-dwelling hard clams are benefited by two successive stages which provide the animal considerable freedom in selecting a definitive habitat. The first, the pediveliger stage, moves by alternation of crawling and swimming with tidal water masses over large areas of bottom within the tidal excursion. Results of investigations of other invertebrate planktonic larvae would suggest that newly settled hard-clam larvae will not affix by means of the byssus within its area of sampling until suitable bottom is found; and in the event that such substrate is not immediately encountered, the larva may possibly postpone metamorphosis and loss of the velum for an extended period (Thorson, 1950). It is not known for *M. mercenaria* how long

metamorphosis may be postponed, or, indeed, to what degree the observed behavior of alternation of crawling and swimming is an indication of "searching." Circumstantial observations suggest that this is so, but further studies should be made to clarify this point. The second phase which provides the larva freedom of choice, though to a much more geographically limited degree, is the byssal stage, in which the animal alternates byssal fixation with crawling on a still highly effective motile foot. Thus in the presence of adverse localized environmental factors the clam can move a short distance and possibly avoid the negative conditions.

On the bottom the young clam meets several problems: displacement by currents once it has settled in a particular substratum, destruction by predators, procurement of food in the midst of an aquatic dust storm. Observations reported in this paper indicate how effectively these difficulties are resolved and how well adapted young hard clams are to their coastal environment.

Soon after pediveligers drop to the bottom they secrete a byssus, by means of which they anchor themselves in position. Plantigrades, now noticeably negatively photokinetic, affix to hard surfaces overlain by thin layers of sediment when these are available. This provides a stable anchorage, permits execution of the affixation process under cover of the sediment, and makes available a sedimentary blanket into which plantigrades can burrow away from enemies as they grow. Where such hard surfaces are not present, plantigrades affix the byssus to clusters of sand grains. In channels where swift tidal currents prevail, young clams and their anchorages are moved about by the currents, though probably to a limited degree; just how much has not been reported. Some field observations also suggest that under these circumstances young clams may be concentrated by tidal currents. Whether such concentrations are actually the result of passive accumulation of young clams by water currents, or whether pediveligers were originally attracted to these sites, has not been determined experimentally. Both processes may well operate.

As soon as siphons start to emerge, plantigrades burrow more deeply into the sediment, and away from predators. Since the byssus is readily cast off, plantigrades may crawl easily from an unfavorable site and reaffix quickly in a new location. Siphons, in addition to permitting clams to burrow deeply, point away from each other and deflect incoming and outgoing streams of water, thereby reducing the degree to which a clam recycles water through its mantle cavity. Siphons may be extended at least twice the length of the valves, which permits clams to burrow to this depth and still maintain direct communication with overlying water. Tentacles over the inhalant siphon form a screen which blocks the entrance of larger sedimentary particles. This allows clams to burrow more deeply than permitted by the length of the siphons alone, for the tentacular grid excludes sediments but permits the entrance of interstitial sea water and fine food and sedimentary particles. The depth to which clams may burrow beyond the length of the siphons is not known with certainty, but may be correlated with porosity of sediment. The inhalant tentacular screen makes possible the existence of young clams in unstable sandy bottoms, keeping out the rain of grains roiled by tidal currents. The clams' capacity to dig themselves into and out of sediment makes it possible for them to move vertically in adjustment to fluctuating bottom levels.

Finally, secondary shell rings, though not affording any great resistance to locomotion through the sediment because they tend to parallel direction of movement, do probably constitute friction elevations which offer some resistance to any force drawing the clams vertically out of the bottom.

VI SUMMARY

1. Of the 2,890 *M. mercenaria* employed to obtain gametes over a period of six summers, 12 per cent spawned. We obtained the most abundant and consistent spawnings in New York, and the fewest in North Carolina. The bulk of spawning came in the early summer, and it became increasingly difficult to induce spawning as the season advanced. Retardation of spawning by keeping the clams at low temperatures helped circumvent this difficulty. In all three geographic areas, fully spawning, vigorous females which eliminated ova quickly and abundantly produced the most vigorous, rapidly growing larvae with least mortality.

2. Larvae of the hard clam were reared in the laboratory in New Jersey, New York, and for the first time in North Carolina—most successfully in North Carolina. They were maintained

in large plunger jars, in large still-water culture jars, and individually in microculture dishes. They survived gradual temperature fluctuations ranging between 13°C. and 30°C. A variety of foods including both cultures of mixed micro-organisms dominated by *Chlorella* and by *Nannochloris* and extracts of pablum were provided. The growth rate of larvae was maximal in sea water hauled from the bay and decreased (in progressive reduction) in sea water pumped into the laboratory, in heat-treated sea water, and in charcoal-treated sea water. Lack of uniformity in culturing larvae under apparently similar conditions within single and in different seasons and in different regions suggests that an unidentified substance(s) of sporadic distribution in native waters may be correlated with success in culturing them. The minimum time for development of the larvae to setting stage was eight days. The mortality of the larvae in cultures was highly variable in time and in place.

3. Early stages of *M. mercenaria* pass through a series of recognizable stages characterized by distinct structural features and behavior: non-shelled planktonic stage, straight-hinged veliger, umboned veliger, prodissoconch I, prodissoconch II (veliconcha stage), pediveliger, byssal plantigrade, and juvenile plantigrade. Position and behavior of these stages in the native habitat change with each successive major morphological development.

4. Veliger stages are characterized by smooth valves (prodissoconchs I and II). These are succeeded in the byssal plantigrade first by primary and then by secondary shell ridges. The latter are added throughout the life of the clam. Because of the curvature of these ridges and the behavior of clams in burrowing, the ridges afford a minimum of resistance to locomotion in sediment, but once the animal has become stationary, they aid in anchoring the animal within sediment.

5. Pedal locomotion in pediveligers and newly set plantigrades involves principally ciliary activity; this is superseded by muscular crawling, which permits the animal to burrow within sediment and to emerge from deep sediment when so buried by pushing itself upward with the foot.

6. Shortly after larvae assume the plantigrade habit, and sometimes before the velum is lost, the byssus gland becomes functional and the bivalve begins attachment to substrata by means of the byssus. The byssus increases in size as the animal grows until it reaches shell length of about 9 mm., at which time the byssus gland becomes functionless. The clam makes attachment to an open surface, but attaches more quickly and more frequently when buried under sediment. Affixation of the byssus takes place in a few seconds. It may be dislodged from its origin in the gland when the animal is disturbed and moves on to another site, and dislodgment is performed less readily as the clam increases in size. In the laboratory, the interval between discard and secretion of a new byssus ranged from a few minutes to several hours in different individuals.

7. In veliger and newly set plantigrade clams, sea water moves through the mantle cavity between gaping valves. Soon tissue union of the mantle edges divides the gape into an exhalant and combination pedal-inhalant opening. From the rim of the exhalant opening, there emerges a valvular membrane as the clam develops which controls and directs the exhalant current. At the base of the membrane there next forms the definitive exhalant siphon with its terminal tentacles, which supports the membrane on its outer extremity as it lengthens. The tentaculated inhalant siphon develops as an extension of the mantle edge next to and anteroventrad the exhalant siphon and from the posterior part of the combination inhalant-pedal gape. As the clam reaches adulthood, most of the valvular membrane is resorbed and is represented only by a reduced flange. Siphon tips point away from each other, deflecting incurrent and excurrent water.

8. When byssal plantigrades are buried deep in sediment, tentacles over the siphon tips form protective grids which exclude larger sedimentary grains. These are cleared by slight forceful appression of the valves. As they crawl within sediment, clams continue to pump water, though at a reduced rate probably corresponding to porosity of the overlying sediment. They can emerge from sediment through a depth at least five times the shell length.

9. In the laboratory, trochophore and early straight-hinged veligers are strong swimmers and tend to concentrate in surface layers of water in culture jars. Later-stage veligers remained more or less evenly distributed vertically. Healthy veligers stayed suspended in the water the full period of planktonic existence, quickly returning to suspension if knocked to the bottom, whereas "sick" larvae remained on the bottom except for sporadic swimming efforts.

10. The behavior and ecology of veligers in native waters was studied in Little Egg Harbor, N. J., a shallow coastal plain estuary with a

water-surface area of about 32 square miles. Float and current studies disclosed that in the central basin of Lower Little Egg Harbor the tidal excursion was relatively short as compared with that on the eastern side. Computations of net movements of fresh water into the bay during the summer indicated that slight flushing of bay water into the ocean fluctuated with net flow of sea water into the bay during periods when evaporation rate exceeded rate of fresh-water inflow. The movement of relatively small volumes of fresh water into the bay from all sources, combined with a complex circulation pattern and wind action, produced relatively uniform, high summer salinities throughout the bay and almost eliminated vertical salinity and thermal stratification. Calculation of volume of water in the bay at various stages of the tide showed that exchange was substantial, but because of the unique morphometry and large size of the bay relative to the large cross section and length of the inlet, and with little or no flushing, the mass of water involved in exchange was principally that in the inlet. Thus the central basin of Lower Little Egg Harbor functioned as a retaining vessel of relatively homogeneous sea water in which a large proportion of clam larvae in many instances were held for at least the duration of the veliger stage.

11. Little Egg Harbor has produced high concentrations of hard clams. In the period 1929–1951 estimates of annual yield from the bay ranged from 40,000,000 to 160,000,000 clams.

12. The distribution of hard-clam veligers was studied in the bay over a period of four summers, during which time 1,100 plankton samples were collected. Each summer the largest and most dense swarms appeared during the month of July, and the number and size of spawnings varied from summer to summer. Most major spawnings occurred in a period of two or more days of rising median daily water temperatures: 73 per cent of a total of 67 spawnings took place on rising temperatures, and 27 per cent on falling median daily temperatures; the over-all average median daily spawning temperature was 25.7°C. Spawnings occurred over a median daily range of water temperatures of 22° to 30°C., but the maximum frequency of spawning occurred within the thermal range of 24°–26°C.

13. Spawnings occurred more frequently and more high density swarms of early-stage veligers appeared in the water during neap than during spring tide periods. Higher water temperatures of neap tides probably triggered spawning during this period. Spawning probably took place at or close to the ebb slack period when maximal water temperatures prevailed.

14. When the appearance of veliger swarms in the bay coincided with periods of little or no rainfall and medium to low tidal amplitudes, and thus minimal flushing and exchange, survival rates of veligers from the straight-hinged to the setting stage approximated 2.6 per cent. Conversely, during periods of heavy rainfall and high spring tidal exchanges even very large dense swarms of larvae disappeared from the water.

15. In Little Egg Harbor at median daily water temperatures ranging from 23.4° to 26.2°C. and in salinities fluctuating between 30.4 and 31.4 °/oo hard-clam veligers grew to a setting size of about 200 μ from time of fertilization in eight days, an interval comparable with that in the laboratory.

16. Horizontal distribution of veligers in Little Egg Harbor was markedly uneven, though widespread. Maximum densities were found in the lower part of the central basin of the bay; the highest single concentration of straight-hinged veligers encountered there was 67,200 per 100 liters of sea water. In the vicinity of the inlet low densities resulted in part from diluting effect of tidal exchange.

17. For the study of vertical distribution of veligers in the bay, a total of 97 vertical series of plankton samples was collected, and the vertical distribution of straight-hinged and umboned veligers was associated with the stage of the tidal cycle, time of day, relative wind velocity, relative light intensity, and current velocity, and location of various stations in the bay. Veligers appear to maintain themselves suspended in the water throughout planktonic existence. They are generally away from the bottom and bottom-dwelling predators during daylight hours, concentrating in middle strata, and more broadly distributed during hours of darkness, distribution of umboned stages especially extending close to the bottom. Light and turbulence from tidal currents and wave action may stimulate larvae to rise vertically, bringing maximum densities higher in the water during maximal tidal flow and wind velocities and during daylight hours. This needs confirmation. Vertical homogeneity of the water with reference to temperature and salinity precluded association of these factors with vertical distribution of the larvae. Early-stage larvae tended to remain in the original swarms resulting from mass spawning by par-

ents, and in time as a result of hydrographic mixing processes late-stage larvae, apparently unable to counteract horizontal dispersion, became widely and more or less uniformly distributed.

18. Behavior of early byssal plantigrades was studied in the laboratory. They are strongly thigmokinetic and attach the byssus most frequently to hard surfaces covered with thin layers of fine sediment containing appreciable quantities of organic matter; they are also negatively photokinetic, crawling away from light of bright to moderate intensity into areas of dim light. No rheotactic response was elicited in current velocities up to 2.5 cm./sec.

19. Sets of hard clams occurred widely though irregularly througout Little Egg Harbor, and their distribution did not coincide entirely with that of adults. The maximum concentration of byssal plantigrades, 125 per square meter, was located on a shell bar where byssal attachment was made to shells coated with thin layers of mud, confirming similar observations in the laboratory. In the field young clams were often found concentrated most densely in sediment around objects projecting from the bottom. The hypothesis is that this results because: (a) pediveligers may be stimulated to set beneath areas of turbulence differentials, (b) they may crawl or swim into the vicinity of such baffles secondarily after settling elsewhere, and (c) baffle areas provide microhabitats which provide some protection form predators. The response of pediveligers to gradients of turbulence would also stimulate them to settle along edges of channels and sloping portions of bars where they may occur in high densities.

20. A small portable device, the clam-setting indicator, was evolved to take advantage of those characteristics in the setting behavior of pediveligers which stimulate them to affix in indicators and provide a means of determining the relative intensity of setting in the field. These characteristics are (a) possible attraction to gradients of turbulence about objects in the water, (b) preference during daylight for microhabitats in subdued light, (c) preference for contact with shallow layers of fine sediment over hard surfaces. The device excludes predators; does not impede flow of water, food, and oxygen; and permits study of plantigrades within it in a minimal quantity of fine sediment. Maximum density, 202 clams per 0.2 square foot, was obtained in a setting indicator near the bottom in the middle of Little Egg Harbor. Intensity of set was roughly proportional to abundance of late-stage veligers in the plankton preceding the set.

21. Enemies of early stages of the hard clam are described. These include wide variety of plankton feeders, *Sirolpidium*, *Condylostoma*, xanthid mud crabs, *Callinectes*, *Carcinides*, *Limulus*, *Urosalpinx*, *Eupleura*, *Polinices*, *Molgula*, *Sphaeroides*, ducks and geese, and possibly *Busycon* and *Asterias*.

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LITERATURE CITED

ANSELL, A. D. 1960. Observations on predation of *Venus striatula* (Da Costa) by *Natica alderi* (Forbes). Proc. Malacolog. Soc. London **34**: 157–164.

———. 1961. Reproduction, growth and mortality of *Venus striatula* (Da Costa) in Kames Bay, Millport. Jour. Mar. Biol. Assoc. U. K. **41**: 191–215.

BAGGERMAN, B. 1953. Spatfall and transport of *Cardium edule* L. Arch. Neerland. Zool. **10**: 315–342.

BELDING, D. L. 1912. A Report upon the Quahaug and Oyster Fisheries of Massachusetts, Including the Life History, Growth and Cultivation of the Quahaug. Dept. Conserv., Commonwealth Mass. Wright and Potter Printing Co.: Boston. 134 pp.

BIGELOW, H. B., AND W. C. SCHROEDER. 1953. Fishes of the Gulf of Maine. U. S. Fish & Wildlife Serv. Fish. Bull. **53**: 526–527.

BOUSFIELD, E. L. 1955. Ecological control of the occurrence of barnacles in the Miramichi Estuary. Natl. Mus. Canada Bull. No. 137; Biol. Ser. No. **46**: 1–69.

BROWNE, E. T. 1897. On keeping medusae alive in an aquarium. Jour. Mar. Biol. Assoc. **5**: 176.

BRUCE, J. R., M. KNIGHT, AND M. W. PARKE. 1940. The rearing of oyster larvae on an algal diet. Jour. Mar. Biol. Assoc. **24**: 343.

BUELL, M. F., AND J. E. CANTLON. 1950. A study of two communities of the New Jersey Pine Barrens and a comparison of methods. Ecology **31**: 567–586.

BURKENROAD, M. D. 1957. Intensity of setting of starfish in Long Island Sound in relation to fluctuations of the stock of adult starfish and in the setting of oysters. Ecology **38**: 164–165.

CARRIKER, M. R. 1950a. Notes on the killing and preservation of bivalve larvae in fluids. Nautilus **64**: 14–17.

———. 1950b. Some recent investigations of native bivalve larvae in New Jersey estuaries. Proc. Natl. Shellfish Assoc. **1950**: 69–74.

———. 1951a. Observations on the penetration of tightly closing bivalves by *Busycon* and other predators. Ecology **32**: 73–84.

———. 1951b. Ecological observations on the distribution of oyster larvae in New Jersey estuaries. Ecol. Monogr. **21**: 19–38.

———. 1952. Preliminary studies on the field culture, behavior, and trapping of the larvae of the hard clam *Venus* (*Mercenaria*) *mercenaria* L. Natl. Shellfish. Assoc. Conv. Addresses **1952**: 70–73.

———. 1956. Biology and propagation of young hard clams, *Mercenaria mercenaria*. Jour. Elisha Mitchell Sci. Soc. **72**: 57–60.

———. 1957. Preliminary study of behavior of newly hatched oyster drills, *Urosalpinx cinerea* (Say). Jour. Elisha Mitchell Sci. Soc. **73**: 328–351.

———. 1959. The role of physical and biological factors in the culture of *Crassostrea* and *Mercenaria* in a salt-water pond. Ecol. Monogr. **29**: 219–266.

———. 1961. Use of a mobile floating laboratory for ecological estuarine research. Ward's Nat. Sci. Bull. **34** (2): 3–7, 26–27.

CHANLEY, P. 1958. Survival of some juvenile bivalves in water of low salinity. Proc. Natl. Shellfish. Assoc. **48**: 52–65.

CHESTNUT, A. F. 1952. Growth rates and movements of hard clams, *Venus mercenaria*. Proc. Gulf Carib. Fish. Inst. 4th Ann. Sess. 49–59.

COAST AND GEODETIC SURVEY. 1941. Manual of Tide Observations. Spec. Publ. No. 196, Rev. ed., U. S. Gov't. Printing Office, Wash. 92 pp.

COLE, H. A. 1938. The fate of the larval organs in the metamorphosis of *Ostrea edulis*. Jour. Mar. Biol. Assoc. U. K. **22**: 469–488.

DAVIS, H. C. 1958. Survival and growth of clam and oyster larvae at different salinities. Biol. Bull. **114**: 296–307.

DAVIS, H. C., AND P. E. CHANLEY. 1956. Effects of some dissolved substances on bivalve larvae. Proc. Natl. Shellfish. Assoc. **46**: 59–74.

DAVIS, H. C., AND R. R. GUILLARD. 1958. Relative value of ten genera of microorganisms as foods for oyster and clam larvae. U. S. Fish & Wildlife Serv. Fish. Bull. **58**: 291–304.

DAVIS, H. C., V. L. LOOSANOFF, W. H. WESTON,

AND C. MARTIN. 1954. A fungus disease in clam and oyster larvae. Science **120**: 36–38.

DE BLOK, J. W., AND HANNIE J. GEELEN. 1958. The substratum required for the settling of mussels (*Mytilus edulis* L.). Arch. Neerland. Zool., Suppl. **1** to **13**: 446–460.

DEW, BARBARA, AND E. J. WOOD. 1955. Observations on periodicity in marine invertebrates. Australian Jour. Mar. & Freshwater Res. **6**: 469–478.

FEEN, P. J. VAN DER. 1949. Byssus. Basteria **13**: 66–71.

FRISSELL, D. L. 1936. Primary reclassification of veneracean pelecypods. Bull. Mus. Roy. d'Hist. Nat. Belgique **12** (34): 1–84.

GAARDER, T., AND P. BJERKAN. 1934. Østers og Østerskultur i Norge. A. S. John Grieg, Bergen.

HANKS, J. E. 1960. The Early Life history of the New England Clam Drills, *Polinices duplicatus* (Say), *Polinices heros* (Say), and *Polinices triseriata* (Say) (Naticidae: Gastropoda). Unpublished Ph.D. dissertation, University of New Hampshire. 122 pp.

HULBERT, E. M. 1957. The distribution of *Neomysis americana* in the estuary of the Delaware River. Limnol. & Oceanogr. **2**: 1–11.

HUNTER, W. R. 1949. The structure and behavior of *"Hiatella gallicana"* (Lamarck) and *"H. arctica"* (L.), with special reference to the boring habit. Proc. Roy. Soc. Edinburgh Sec. B. **63** (3) (19): 271–289.

JENNER, C. E. 1956. The occurrence of a crystalline style in the marine snail, *Nassarius obsoletus*. Biol. Bull. **111**: 304 (abstract).

JØRGENSEN, C. B. 1946. Lamellibranchia *in* Reproduction and larval development of Danish marine bottom invertebrates by G. Thorson. Medd. Komm. Danmarks Fish. Havunders. Ser. Plankton **4**: 277–311.

KELLOGG, J. L. 1892. A contribution to our knowledge of the morphology of lamellibranchiate mollusks. Bull. U. S. Fish. Comm. **10** (for 1890): 389–436.

———. 1901. Clam and scallop industries of New York State. Bull. N. Y. Sta. Mus. No. 43, vol. **8**: 605–629.

KORRINGA, P. 1940. Experiments and observations on swarming, pelagic life and setting in the European flat oyster, *Ostrea edulis* L. Arch. Neerland. Zool. **5**: 1–249.

LANDERS, W. S. 1954. Seasonal Abundance of Clam Larvae in Rhode Island Waters, 1950–52. U. S. Fish & Wildlife Serv. Spec. Sci. Rept.—Fish. No. 117.

LEBOUR, M. V. 1933. Rearing marine animals in a plunger. Sci. Progr. **107**: 494–502.

LINDSAY, C. E., AND C. E. WOELKE. 1960. Production of clam and oyster seed. In Fisheries, Vol. III, by M. Moore, K. McLeod, and D. Reed (Wash. Sta. Dept. Fish.), pp. 81–85.

LOOSANOFF, V. L. 1937a. Development of the primary gonad and sexual phases in *Venus mercenaria* Linnaeus. Biol. Bull. **72**: 389–405.

———. 1937b. Spawning of *Venus mercenaria* (L.). Ecology **18**: 506–515.

———. 1953. Lack of relation between age of oysters or clams and quality of their spawn. U. S. Fish & Wildlife Fish. Biol. Lab., Milford, Conn., Bull. **17** (4): 1–2.

———. 1954. New advances in the study of bivalve larvae. Amer. Sci. **42**: 607–624.

———. 1958. Challenging problems in shellfish biology. In Perspectives in Marine Biology, A. A. Buzzati-Traverso, ed. Berkeley and Los Angeles: University of California Press.

———. 1959a. *Condylostoma*—an enemy of bivalve larvae. Science **129**: 147.

———. 1959b. The size and shape of metamorphosing larvae of *Venus* (*Mercenaria*) *mercenaria* grown at different temperatures. Biol. Bull. **117**: 308–318.

LOOSANOFF, V. L., AND H. C. DAVIS. 1950. Conditioning of *V. mercenaria* for spawning in winter and breeding its larvae in the laboratory. Biol. Bull. **98**: 60–65.

——— AND ———. 1951a. Delayed spawning of lamellibranchs by low temperature. Jour. Mar. Res. **10**: 197–202.

LOOSANOFF, V. L., H. DAVIS, AND P. CHANLEY. 1953. Effect of overcrowding on rate of growth of clam larvae. Anat. Rec. **117**: 645 (abstract).

———, ———, AND ———. 1953b. Behavior of clam larvae in different concentrations of food organisms. Anat. Rec. **117**: 586 (abstract).

LOOSANOFF, V. L., AND J. B. ENGLE. 1942. Use of complete fertilizers in cultivation of microorganisms. Science **95**: 487–488.

LOOSANOFF, V. L., J. E. HANKS, AND A. E. GANAROS. 1957. Control of certain forms in zooplankton in mass algal cultures. Science **125** (3257): 1092–93.

LOOSANOFF, V. L., W. S. MILLER, AND P. B. SMITH. 1951b. Growth and setting of larvae of *Venus mercenaria* in relation to temperature. Sears Found. Jour. Mar. Sci. **10**: 59–81.

LYNCH, W. F. 1959. Factors influencing metamorphosis of larvae of some of the sessile organisms. XVth Internatl. Congr. Zool. London, Proc.: 239–241.

MANNING, J. H., AND H. H. WHALEY. 1954. Distribution of oyster larvae and spat in relation to some environmental factors in a tidal estuary. Proc. Natl. Shellfish. Assoc. **45**: 56–65.

MARTIN, A. C., AND F. M. UHLER. 1939. Food of Game Ducks in the United States and Canada. U. S. Dept. Agr. Tech. Bull. 634. 157 pp.

MORTON, J. E. 1960. The responses and orientation of the bivalve *Lassaea rubra* Montagu. Jour. Mar. Biol. Assoc. U. K. **39**: 5–26.

NELSON, T. C. 1921. Aids to successful oyster culture. I. Procuring the seed. N. J. Agr. Exper. Sta. Bull. **351**: 4–59.

NELSON, T. C., AND H. H. HASKIN. 1949. On the spawning behavior of oysters and of *Venus mercenaria* with especial reference to the effects of spermatic hormones. Anat. Rec. **105**: 484–485 (abstract).

NELSON, T. C., AND E. B. PERKINS. 1931. Annual Report of the Department of Biology for the year ending June 30, 1930. N. J. Agr. Exper. Sta.

ORTON, J. H. 1937. Some interrelations between bivalve spatfalls, hydrography and fisheries. Nature **140**: 505–506.

POMEROY, L. R., H. H. HASKIN, R. A. RAGOTZKIE. 1956. Observations on dinoflagellate blooms. Limnol. & Oceanogr. **1**: 54–60.

PRITCHARD, D. W. 1951. The physical hydrography of estuaries and some applications to biological problems. Trans. 16th North Amer. Wildlf. Conf., Wildlife Management Institute: 368–376.

———. 1952. Salinity distribution and circulation in the Chesapeake Bay estuarine system. Jour. Mar. Res. **11**: 106–123.

———. 1955. Estuarine circulation patterns. Proc. Amer. Soc. Civil Engineers **81**: 1–11.

PRITCHARD, D. W., AND R. E. BUNCE. 1959. Physical and Chemical Hydrography of the Magothy River. Chesapeake Bay Institute, Johns Hopkins Univ., Tech. Rept. 17.

QUAYLE, D. B. 1949. Movements in *Venerupis* (*Paphia*) *pullastra* (Montagu). Proc. Malacol. Soc. London **28**: 31–37.

———. 1952. Structure and biology of the larva and spat of "*Venerupis pullastra*" (Montagu). Trans. Roy Soc. Edinburgh **62** (1) (8): 255–297.

RAGOTZKIE, R. A. AND R. A. BRYSON. 1953. Correlation of currents with the distribution of adult *Daphnia* in Lake Mendota. Jour. Mar. Res. **12**: 157–172.

RAVEN, C. P. 1958. Morphogenesis: The Analysis of Molluscan Development. New York: Pergamon Press.

REES, C. B. 1950. The identification and classification of lamellibranch larvae. Hull Bull. Mar. Ecol. **3**: 7–103.

RICE, T. R. 1953. Phosphorus exchange in marine phytoplankton. U. S. Fish & Wildl. Serv. Fish. Bull. **54**: 76–89.

RICE, T. R., AND REBECCA J. SMITH. 1958. Filtering rates of the hard clam (*Venus mercenaria*) determined with radioactive phytoplankton. Fish. Bull. U. S. Fish. & Wildlife Serv. **58**: 73–82.

RICHARDS, O. W. 1933. Toxicity of some metals and Berkefeld filtered sea water to *Mytilus edulis*. Biol. Bull. **65**: 371–372.

RYDER, J. A. 1889. The byssus of the young of the common clam *Mya arenaria* L. Amer. Nat. **23**: 65–67.

SAWYER, D. B. 1950. Feeding activities of the boring snail *Polynices duplicata*. 3rd Rept. Invest. Methods Improving Shellf. Res. Mass., Commonwealth of Mass., Dept. Conserv., Div. Mar. Fish.: 16–17.

SCHEFFÉ, H. 1959. The Analysis of Variance. New York: John Wiley & Sons, Inc. Pp. 331-368.

SELLMER, G. P. 1959. Studies on the Anatomy and Life History of the Gem Clam, *Gemma gemma* (Totten). Unpublished Ph.D. dissertation, Rutgers University, New Brunswick, N. J. Pp. 237.

SHUSTER, C. N. 1950. Observations on the natural history of the American horseshoe crab *Limulus polyhemus*. 3rd Rept. Invest. Methods Improving Shellfish. Res. Mass. Commonwealth of Mass. Dept. Conserv., Div. Mar. Fish.: 18–23.

———. 1951. On the migration of young *Polynices*. Anat. Rec. **111**: 542 (abstract).

STAFFORD, J. 1912. On the recognition of bivalve larvae in plankton collections. Contr. Canadian Biol. 1906–10: 221–242.

SULLIVAN, C. M. 1948. Bivalve larvae of Malpeque Bay, P.E.I. Fish. Res. Bd. Canada Bull. **78**: 1–36.

SVERDRUP, H. V., M. W. JOHNSON, AND R. H. FLEMING. 1942. The Oceans. Their Physics, Chemistry, and General Biology. New York: Prentice-Hall, Inc.

THORSON, G. 1946. Reproduction and larval development of Danish marine bottom invertebrates, with special reference to the planktonic larvae in the sound (Øresund). Medd. Komm. Danmarks Fisk. Havunders. Ser. Plankton **4**: 1–523.

———. 1950. Reproductive and larval ecology of marine bottom invertebrates. Biol. Rev. Cambridge **23**: 1–45.

———. 1957. Bottom communities. Chapt. 17 in Treatise on Marine Ecology and Paleoecology, vol. 1, memoir 67, Geol. Soc. Amer.: 461–534.

TURNER, H. J. 1953. A review of the biology of some commercial molluscs of the east coast of North America. 6th Rept. Invest. Shellf. Mass., Dept. Nat. Res., Div. Mar. Fish., Commonwealth Mass.: 39–74.

TURNER, H. J., AND C. J. GEORGE. 1955. Some aspects of the behavior of the quahaug, *Venus mercenaria*, during the early stages. 8th Rept. Invest. Shellf. Mass., Dept. Nat. Res., Div. Mar. Fish., Commonwealth Mass.: 5–14.

U. S. GEOLOGICAL SURVEY. 1948–1951. Surface Water Supply of the United States. Part I. North Atlantic Slope Basins. Water-supply Papers 1111, 1141, 1171, 1202. U. S. Geol. Surv., U. S. Dept. Int., U. S. Gov't. Printing Office, Wash.

U. S. WEATHER BUREAU. 1948–1951. Climatological data, New Jersey. U. S. Weather Bur., U. S. Dept. Commerce.

WELLS, H. W. 1957. Status of the name *Venus*. Ecology **38**: 160–161.

WELLS, W. F. 1927. Report of experimental shellfish station. 16th Ann. Rept. **1926**: 1–22.

WERNER, B. 1939. Uber die Entwicklung und Artunterscheidung von Muschellarven des Nordseesplanktons, unter besonderer Berucksichtigung der Schalenentwicklung. Zool. Jahrb., Abt. Anat. **66**: 1–54.

WINSOR, C. P. AND G. L. CLARK. 1940. A study of variation in the catch of plankton nets. Jour. Mar. Res. **3**: 1–34.

YONGE, C. M. 1957. Mantle fusion in the lamellibranchia. Pubbl. Staz. Zool. Napoli **26**: 151–171.

A *PALAVASCIA* (ECCRINALES) FROM THE MARINE ISOPOD *SPHAEROMA QUADRIDENTATUM* SAY

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A new species of fungus, belonging to the interesting genus *Palavascia*, has been found living within the hind-gut of the small intertidal marine isopod *Sphaeroma quadridentatum* Say. This is the first species of *Palavascia* reported in the Western Hemisphere. It belongs to the Trichomycete order Eccrinales, all species of which live as obligate commensals attached to the chitinous gut lining of arthropods.

The present paper describes and illustrates the new fungus, and discusses some biological relationships of the two organisms.

Materials and Methods

This study is based upon a single population of *Sphaeroma* living in a restricted area known locally as "the old sluiceway," about 250 meters west of the Piver's Island bridge, which leads to the Duke University Marine Laboratory, between Morehead City and Beaufort, North Carolina.

The isopods were found living in the intertidal zone among rocks and debris piled against a decaying wood wall and in the crevices of the wood itself. Although the habitat was more or less restricted, the population was large and prolific, and repeated collecting did not cause any great reduction in the population.

Observations and collections were made when the tide was sufficiently low to expose the beach. Under these conditions, the *Sphaeroma* congregated in places that remained moist until high tide, such as underneath rocks and seaweed, within dead barnacle shells, or in the cracks and laminations present in the decaying wood.

Collections were made personally in July and August of 1958, and again in June of 1959. An additional collection was made in November of 1958 by Dr. F. John Vernberg of the Duke University Marine Laboratory, who shipped the material to me at The University of Kansas.

The *Sphaeroma* collected were examined and dissected immediately or they were kept alive in containers for varying periods of time. One collection has been kept in our laboratory for over eighteen months, having gone through many reproductive cycles. *Sphaeroma quadridentatum* can be cultured without much difficulty in aquaria with aerated natural sea water, though artificial sea water also can be used. The food used has been rolled oats, fed daily in small quantities.

Dissection and preparation of slides have been carried out essentially as described previously for other arthropods (Lichtwardt, 1954). The living *Sphaeroma* is held down forcibly while the posterior pleopods are spread or torn off, and the hind-gut is pulled out. The gut is cut open with a scalpel made from a minuten insect needle. The lining is peeled from the epithelial tissue, or, if this is difficult, the gut first is soaked in dilute lactophenol (1 drop per 10 ml. water) for a few hours. The final mount is made in lactophenol containing cotton blue.

Slides for primarily cytological studies were prepared by using a propriono-carmine stain technique, although the results were not always satisfactory due to the affinity of the cytoplasm for the stain, especially in sporulating portions of the thalli.

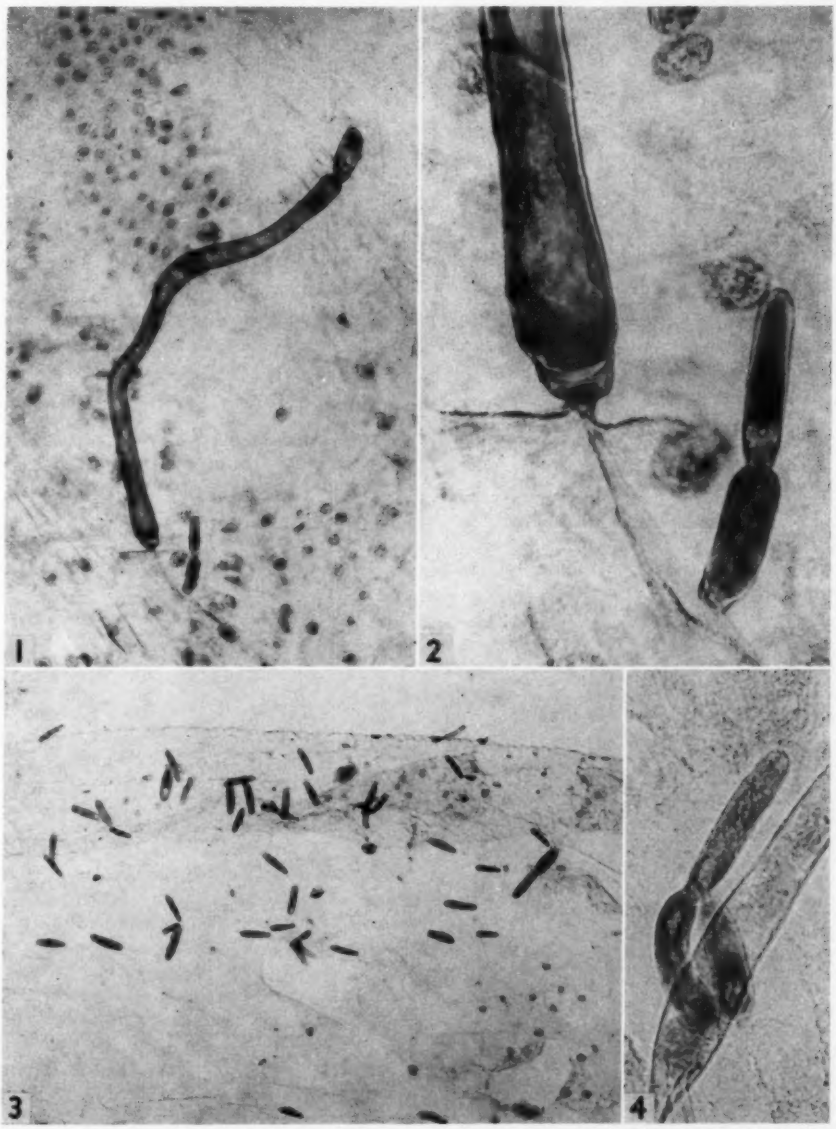
Observations

Palavascia beaufortae sp. nov.

Thallis sterilibus sinuatis, diam. non aequalibus cum incremento basali, lato retinaculo brevi adjunctis, cum maternis sporis persistentibus apice. Thallis maturis propinquis basi circa 30–60 μ *diam., propinquis apice circa* 12–30 μ *diam., usque ad* 6 *mm. long. Apicibus fertilibus spiralibus, serialibus terminalibus cellulis producentibus sporangiosporas solitarias multinucleates ellipsoidas cummuris levibus et crassis,* 29–34 × 15–22 μ, *aut germinatibus in situ et producentibus unam aut plures hyphas laterales, usque ad* 170 μ *long., propinquas basi* 5–10 μ *lat. et propinquas apicibus* 3–4 μ *lat., parte terminali dividenti in seriales cellulis parvis.*

Vegetative thalli sinuate, somewhat uneven in diameter, basal portion becoming enlarged, attached basally by means of a short, blunt holdfast, with the mother-spores remaining attached

apically throughout their development. Mature thalli about 30–60 μ diameter near the base and 12–30 μ diameter near the apex, up to about 6 mm. in length. Sporulating tips coiled, consisting of a long series of cells that either develop into single, thick-walled, smooth, multinucleate, ellipsoid sporangiospores measuring 29–34 × 15–22 μ, or that germinate *in situ*, each producing one or more lateral hyphae, up to about 170 μ long by 5–10 μ wide near the base



FIGS. 1–4. *Palavascia beaufortae*. 1. A young thallus and a germinating spore attached to the hind-gut lining of *Sphaeroma quadridentatum*, X100. 2. Enlarged view of holdfasts, X430. 3. Portion of the gut lining of a very small *Sphaeroma* that contained about sixty ingested and attached spores, one of which is seen germinating, X100. 4. An abnormal development in which one spore is germinating to produce two hyphae, X430.

and 3–4 μ wide near the apex, with a terminal series of small cells.

Attached to the hind-gut lining of the isopod *Sphaeroma quadridentatum* Say. R. W. Lichtwardt slide DML–19–11, TYPE.

Palavascia beaufortae begins its development in a host when a spore is ingested and reaches the anterior or middle part of the hind-gut. A small holdfast develops at one end of the spore, and it becomes firmly anchored to the nonliving, chitinous material that lines the gut. The thallus arises by growth between the holdfast and the spore, with the mother-spore remaining apically attached and alive throughout the growth of the thallus (Figs. 1 and 2), until the reproductive phase is initiated, at which time the mother-spore is delimited by a septum from the thallus proper and disintegrates.

The vegetative thallus is unbranched, nonseptate, coenocytic, and characteristically sinuate and uneven in diameter, particularly as it matures. The basal portion usually swells to two or three times the average diameter of the rest of the filament. Occasional thalli may be seen swollen near the tip (one abnormal hypha I saw measured 100 μ just posteriad to the tip).

The nuclei are small and very numerous. In properly stained material, hundreds of nuclei measuring 3–5 μ can be seen located in the predominantly peripheral cytoplasm.

An interesting and important feature in *P. beaufortae* development is not to be found in descriptions of other species of *Palavascia*, though perhaps it does occur in other species as well. The thalli of *P. beaufortae* do not sporulate until the tips reach the vicinity of the anus of the isopod. Sporulation occurs in the rectum just inside the anus, or the helical tips may actually protrude from the anus in many specimens. The importance of this phenomenon is, first, that the sporulating tips apparently are not harmed by exposure to the external salt-water environment, and, second, the spores produced in the tips do not infect the same individual host. As a consequence, each thallus that develops in a host has an exogenous origin. Further comments on this matter will be made later.

When the vegetative filament reaches the posterior end of the hind-gut, and as sporulation and further growth proceed, the tip becomes incurved, forming a series of coils whose axis is at right angles to the axis of the filament (Fig. 5). Sporulation is preceded by an increase in density of the protoplasm at the distal end of the filament, followed by septation beginning terminally and proceeding basally until dozens of cells are formed. While this is happening, the mother-spore disintegrates, and the older terminal cells mature.

The cells thus formed are sporangia. However, in *Palavascia* the sporangia may develop in two ways. The first, and major, development is the formation of a series of solitary sporangiospores (Fig. 6). In *P. beaufortae* they are smooth walled, ellipsoid, and eight-nucleate at maturity. Though the walls are relatively thick (2–3 μ), they do not appear to be very rigid. As far as I can determine, the sporangiospores arise asexually, though Tuzet and Manier (1948; Manier, 1950) describe in their species what they consider to be a sexual process involved in the formation of the spores. This will be discussed later.

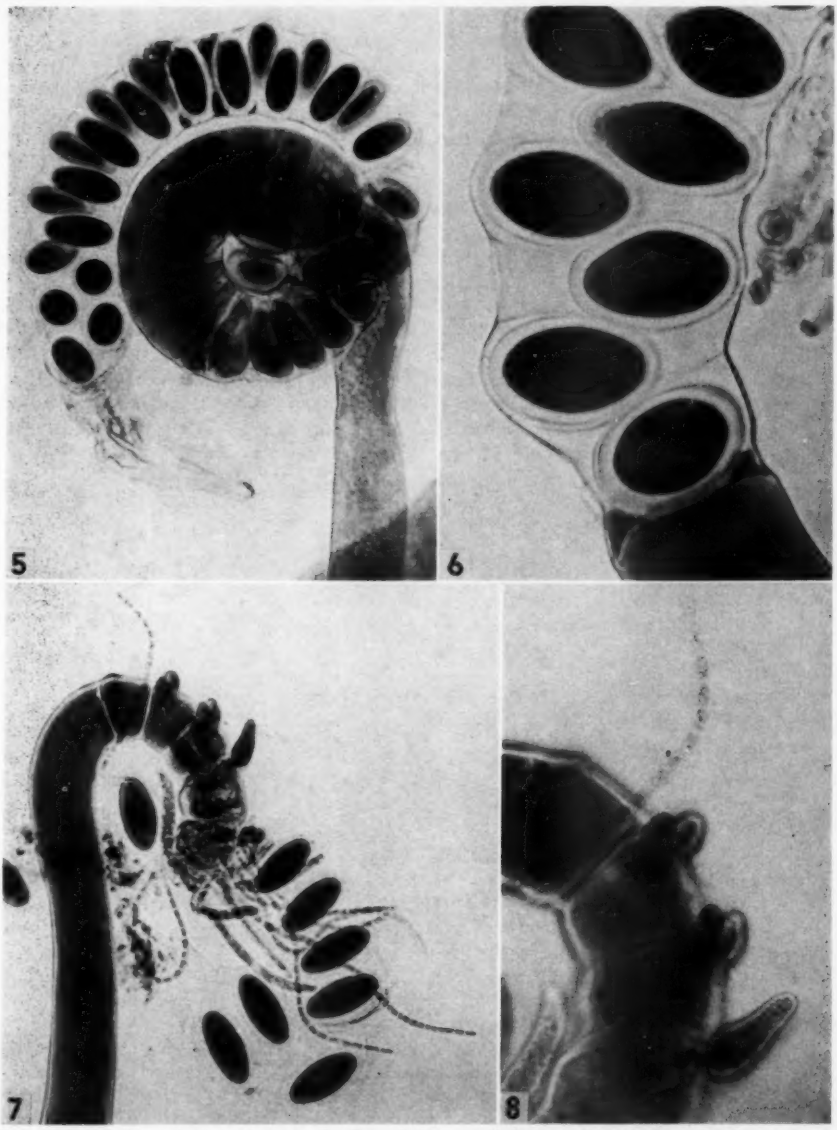
The second type of development of the sporangia, if they do not produce sporangiospoeres, is their germination *in situ*, sending out one to several lateral branches (Figs. 7 and 8). These begin as small buds that swell and become pyriform (at this state reminiscent in shape of the lateral conidia of the branched Trichomycete *Spartiella barbata* Tuzet and Manier in the Genistellales), then elongate into filaments, perhaps over 100 μ long. The terminal portion eventually divides into a series of small cells, starting with the apex and progressing downward. I have not been able to attribute any function to these very small cells, and I can see no way in which they fit into the necessary reproductive cycle of the fungus.

When slides are examined, individual thallus tips may show only sporangiospores, only germinating sporangia, or a combination of both. If only germinating sporangia are to be found in a hyphal tip, it is usually because sporangiospores already have been produced and shed. It is not uncommon to find abortive cells interspersed among the sporangia.

In one instance I was able to watch under the microscope the production of lateral branches from a sporangium mounted in sea water. When first observed, the sporangium had two small buds; within a period of twelve hours these grew to filaments measuring 70 and 85 μ respectively, and a third bud appeared and grew to 25 μ in length. No further development ensued.

Biological Relationships of Host and Fungus

In the genus *Enterobryus* and other genera of the Eccrinaceae there are two principal types of sporangiospores in the life cycle of



FIGS. 5 8. *Palavascia beaufortae*. 5. Sporulating, coiled tip of a thallus showing a series of sporangiospores and sporangia, with an empty, terminal mother-spore, X430. 6. Sporangiospores separated by fine sporangial walls, X970. 7. Tip of a thallus with sporangiospores and lateral hyphae produced by germinating sporangia, X430. 8. Enlargement of germinating sporangia and one lateral hypha tip with a series of very small terminal cells, X970.

the fungi. One spore is multinucleate (usually four or eight nuclei), is more or less cylindrical, and germinates in the same gut in which it was produced. By this means, a host with a single established thallus can eventually become endogenously infected with scores of new thalli. The second important type of sporangiospore in the Eccrinaceae is uninucleate, usually ellipsoid, and sometimes thick walled. It seems to germinate only after it has been eliminated

by one host and ingested by another susceptible individual.

In the genus *Palavascia* (Palavasciaceae) only one principal type of sporangiospore is produced. It functions in the same manner as do the uninucleate spores of the Eccrinaceae, but morphologically it is a combination of the two types, being ellipsoid and thick walled but multinucleate.

As mentioned previously, the sporangiospores of *Palavascia beaufortae* are formed only when the tips of the thalli grow to the region of the anus, and the spores produced do not germinate in the rectum. Yet the fungus can be highly successful in its commensalism, as evidenced by the fact that the population of *Sphaeroma* I studied was virtually 100 per cent infected. Only a very few of the large number dissected did not contain at least some form of the fungus in the gut, and these were among the youngest of the population. Infected isopods supported anywhere from a single thallus to many dozens of thalli in various stages of development. Since infection appears to be entirely exogenous for each thallus in the gut, the success in the passive transfer of spores of *P. beaufortae* must be attributed in large part to the habits of the host.

The fact that even very small specimens of *S. quadridentatum* were found to contain attached spores or thalli in their guts, sometimes in large numbers (Fig. 3), is not too surprising when one observes them in their natural habitat. *S. quadridentatum* appears to be viviparous. The young emerge and become attached to the female's pleopods. Even after they become free-swimming, they often can be found associated with older individuals in the relatively large ventral concavity formed by the telson into which the anus discharges fecal matter. This area remains constantly moist, for if a *Sphaeroma* finds itself in a dry situation, it will roll up into a tight sphere (Fig. 12). By their locating themselves underneath the telson, the small *Sphaeroma* have an excellent and perhaps vital means of preventing desiccation and death at low tide. By these very intimate associations with larger isopods, they are directly exposed to ingestion of the spores which project from or are released from the anus.

The close contact of large *Sphaeroma* at low tide also affords ample opportunity for exchange of inoculum. For instance, several isopods frequently can be found occupying the same barnacle shell or similar partially enclosed habitat, where they remain perhaps for hours until inundated again by sea water.

Each time a *Sphaeroma* molts it sheds the chitinous lining of the hindgut, and along with it all fungal thalli. The intermolt periods of younger individuals are relatively short, and I do not believe that most thalli are capable of reaching the sporulation stage in such cases. Consequently, one finds mature, sporulating filaments commonly only in larger specimens.

Two different cultures of *S. quadridentatum* from Beaufort, N. C., have been maintained in our laboratory at The University of Kansas for experimental purposes. In both cases, after many months of keeping the isopods alive, we found a decrease in the number of individuals infected, and finally all of the isopods seemed to become devoid of the commensal, judging from sample dissections. I attribute this loss of infection primarily to the change in habit of the *Sphaeroma* kept in aquaria, for even though hundreds of the animals were contained in some small aquaria, they were not forced into the same confined situations that are demanded by the changing tides. As they molted and rid themselves of the fungi, their chances of reinfection apparently had decreased.

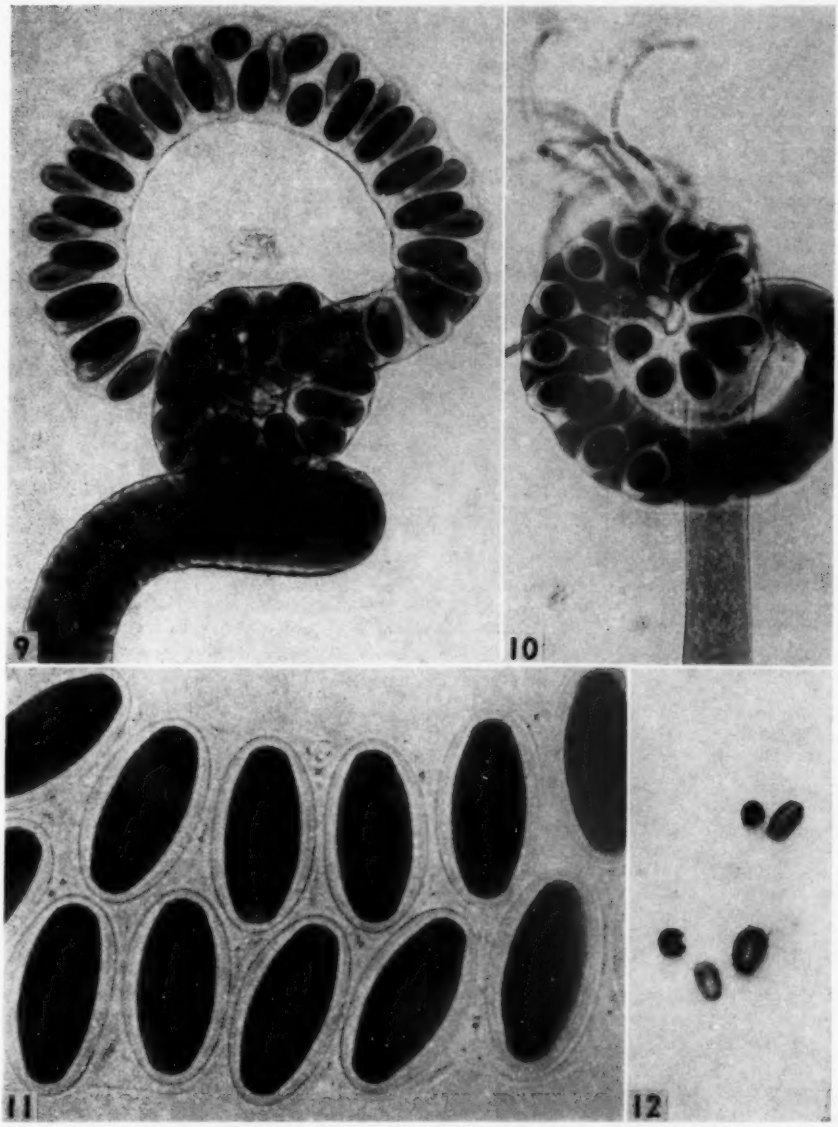
Discussion

Only two other species of *Palavascia* have been described in the literature. Tuzet and Manier established the genus in 1947 with the discovery of *Palavascia philoscii* in the isopod *Philoscia couchii* Kin. The following year (1948) they described the second species, *P. sphaeromae*, found in the isopod *Sphaeroma serratum* F.

The genus *Palavascia* is easily distinguished from other genera of the Trichomycetes. It shows affinities to the order Eccrinales, in which it is placed, because it has unbranched, nonseptate, coenocytic vegetative thalli, attached to the host by a secreted holdfast, and produces a linear series of terminal sporangia. It is placed in a separate family because the genus has sporangia that produce but one major type of sporangiospore, which is multinucleate, ellipsoid, and thick walled; or the sporangia may germinate to produce lateral hyphae that segment into terminal cells.

The diagnostic differences and similarities of the three known species of *Palvascia* can be summarized in the following key:

1. Sporulating tips straight; germinating sporangia terminal, each producing but a single lateral hypha; sporangiospores spiny;



FIGS. 9–12. ***Palavascia beaufortae*** **and host. 9. Portion of a sporulating thallus that had formed about 100 cells and spores, X430. 10. Sporulating tip of a thallus with fertile sporangia alternating with nonfertile ones, X430. 11. Sporangiospores, showing typical shape and thick wall, X970. 12. Living specimens of *Sphaeroma quadridentatum* Say, one completely and another partly curled up, X1.**

found in *Philoscia couchii* Kin. . . . . .
. . . . . *P. philosciae* Tuzet & Manier

1. Sporulating tips coiled; germinating sporangia usually not terminal, each producing one to several lateral hyphae; sporangiospores spiny or smooth . . . . . .

2. Tip producing one coil; lateral branches cylindrical throughout; sporangiospores 4-nucleate at maturity, often spiny; found in *Sphaeroma serratum* F. . . . . *P. sphaeromae* Tuzet & Manier

2. Tips usually producing several coils;

lateral branches usually wider near base; sporangiospores 8-nucleate at maturity, always smooth; found in *Sphaeroma quadridentatum* Say . . . . *P. beaufortae*

Palavascia sphaeromae and *P. beaufortae* are similar in many respects. However, they can be distinguished not only by the above key characters, but also on the basis of a number of other morphological details, among which are the measurements of various structures. The thalli of *P. sphaeromae* attain lengths of 2.5 mm., whereas those of *P. beaufortae* may be as long as 6 mm. The spore dimensions also appear to be different: those of *P. sphaeromae* apparently measure in the neighborhood of 21 μ in length,[1] while those of *P. beaufortae* average 29–34 μ in length.

One very significant difference between the two European species and *P. beaufortae* lies in the development of the sporangiospores as described by Tuzet and Manier and by me. Tuzet and Manier reported in 1948 (and Manier described this in more detail in 1950) that they observed the fusion of adjacent protoplasts in thallus tips undergoing sporulation. This plasmogamy and karyogamy produced cells with single diploid nuclei and was followed soon after by a reduction-division that produced four haploid nuclei in each mature spore.

In my studies of *P. beaufortae* I have not seen such a sequence of developments. The nuclei are small and the cytoplasm is dense in the sporulating tips of thalli (apparently this is the situation in the other species as well). Where nuclei were discernible, the development appeared no different from the asexual formation of uninucleate or multinucleate sporangiospores of *Enterobryus* (Eccrinales) that I have described in other papers (see Lichtwardt, 1954). It remains to be seen whether better preparations will reveal sexuality in *P. beaufortae* development, or whether a difference actually exists between this species and others, or whether Tuzet and Manier have not correctly interpreted the cytological behavior in their two species.

In 1950 Manier established a new class of fungi, the "Palavascides," with the single genus *Palavascia*. This was later changed to an order in the class "Eccrinides" (Tuzet and Manier, 1951, 1953), and finally was reduced to a family of the Eccrinales (Manier, 1955; Tuzet, Manier, and Jolivet, 1957).[2] This last arrangement seems to me to be the correct one, on the basis of our present knowledge of these fungi.

[1] I can find no precise measurements for the sporangiospores of *P. sphaeromae*. The above spore measurement is based upon Manier, 1950, Figure 16. However, Tuzet and Manier (1948) mention that thalli of *P. sphaeromae* measure up to 33 μ in diameter, so that perhaps the spores may be larger than indicated above.

Summary

A new fungus, *Palavascia beaufortae*, is described living as an obligate commensal attached to the hind-gut lining of the intertidal isopod *Sphaeroma quadridentatum* Say, collected near Beaufort, North Carolina. Sporangiospores are produced only in the anal region of the host. The habits of the isopods that insure their survival at low tide also seem to assist in successfully transmitting spores from one individual to another. This species appears to be closely related to *Palavascia sphaeromae* Tuzet and Manier from *Sphaeroma serratum* F. in France.

ACKNOWLEDGMENTS

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I am grateful to Dr. C. G. Bookhout, director of the Duke University Marine Laboratory, for making my studies at Beaufort possible and especially to Dr. F. John Vernberg for his kindness in sending me specimens of *Sphaeroma* and other marine arthropods from the vicinity of Beaufort, N. C.

Dr. Austin Lashbrook, Department of Latin and Greek, The University of Kansas, kindly assisted with the Latin diagnosis.

LITERATURE CITED

LICHTWARDT, R. W. 1954. Three species of Eccrinales inhabiting the hindguts of millipeds, with comments on the Eccrinids as a group. Mycologia **46**: 564–585.

[2] Tuzet and Manier recently have not been following the International Code of Botanical Nomenclature regarding proper suffixes for orders and families. They refer, for example, to the order "Eccrinida" (Eccrinales) and the family "Palavasciidae" (Palavasciaceae).

MANIER, JEHANNE-FRANÇOISE. 1950. Recherches sur les Trichomycètes. Ann. Sci. Nat., Bot., Ser. 11, **11**: 53–162.

———. 1955. Classification et nomenclature des Trichomycètes. Ann. Sci. Nat., Zool., Ser. 11, **17**: 395–397.

TUZET, ODETTE, AND JEHANNE-FRANÇOISE MANIER. 1947. *Palavascia philoscii* n. g. n. sp., Entophyte eccriniforme parasite de *Philoscia Couchii* Kin. C. R. Acad. Sci. **225**: 1854–1856.

——— AND ———. 1948. La reproduction sexuée chez *Palavascia philoscii* Tuzet et Manier et chez *Palavascia sphaeromae*, nouvelle espèce de Palavasciées parasite de *Sphaeroma serratum* F. C. R. Acad. Sci. **226**: 2177–2178.

——— AND ———. 1951. Le cycle de l'*Amoebidium parasiticum* Cienk. Revision du genre *Amoebidium*. Ann. Sci. Nat., Zool., Ser. 11, **13**: 351–364.

——— AND ———. 1953. Recherches sur quelques Trichomycètes rameux. *Asellaria armadillidii* n. sp. *Genistella choanifera* n. sp. *Genistella chironomi* n. sp. *Spartiella barbata* Tuzet et Manier. Ann. Sci. Nat., Zool., Ser. 11, **15**: 373–390.

TUZET, ODETTE, JEHANNE-FRANÇOISE MANIER, AND P. JOLIVET. 1957. Trichomycètes monaxes et rameux de l'intestin postérieur de Polydesmida, Spirostreptida et Spirobolida. Nationaal Albert Park (Ser. 2) **5**: 21–38.

SEASONAL ABUNDANCE OF AQUATIC INSECTS IN WESTERN NORTH CAROLINA TROUT STREAMS

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Introduction

One phase of a Dingell-Johnson project conducted by the North Carolina Wildlife Resources Commission in the Coweeta Experimental Forest in Macon County, North Carolina, was a quantitative and qualitative evaluation of the seasonal variations in standing crops of invertebrate bottom organisms in the forest streams. These data, collected from typical Southern Appalachian trout streams, provide information on seasonal variations of the predominantly insect fauna which should be of interest to students and collectors of aquatic insects of the Southern Appalachians. They should also be of value to aquatic biologists for quantitative comparisons of trout foods during different seasons of the year in streams of this area.

The Coweeta Experimental Forest was established by the United States Forest Service in 1933 for research in watershed management. The forest is located in a mountainous area with elevations ranging from 2,200 to 5,200 feet, and it encompasses an area of 4,000 acres which is approximately evenly divided between two major drainage basins. These two basins are drained by two creeks—Ball Creek on the south and Shope Creek on the north (Fig. 1)—that join within the forest to form Coweeta Creek, which subsequently flows into the Little Tennessee River.

Both Shope and Ball creeks are typical small Southern Appalachian trout streams with very soft, slightly acid water (Table I). They are characterized by steep gradients with series of cascades and low waterfalls interspersed with large pools. The bottom is predominantly rubble and gravel with occasional outcrops of granite bedrock. The average monthly streamflow for a six-year period in Shope Creek ranged from a low 2.31 cfs during October to 8.32 cfs during February. Water temperature on the lower part of these streams reaches a maximum of 70°F. during the month of August.

Over a period of seventeen months a total of 276 samples were collected with a Surber square-foot sampler from riffles at two stations on Ball Creek and one station on Shope Creek. These samples were combined by months to measure the seasonal variations in the fauna.

Total Fauna

The streams of Coweeta Experimental Forest would be classified by most standards as poor producers of bottom organisms. The maximum average monthly volume of 0.86 cc. per square foot was obtained in June, 1954. The maximum volume of bottom organisms in any

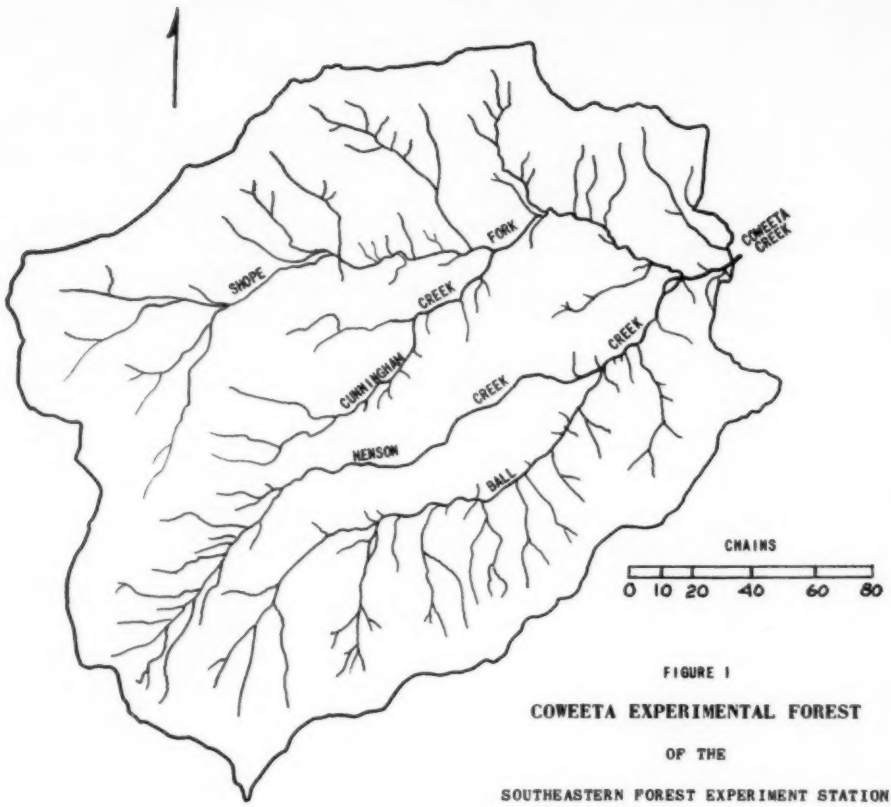


FIGURE 1

COWEETA EXPERIMENTAL FOREST

OF THE

SOUTHEASTERN FOREST EXPERIMENT STATION

Table I

Characteristics of Ball Creek and Shope Creek, Coweeta Experimental Forest, Macon County, N. C.

Characteristics	Shope Creek	Ball Creek
Average channel gradient (ft./mile)	581	644
Drainage area (acres)	1,877	1,788
Aspect	ENE	NE
Elevation at source (feet)	4,000	4,000
*p*H	6.8	6.8
M.O. Alk. ppm.*	8.0	8.0
Phenolphthalein Alk. ppm.*	0.0	0.0
CO_2 ppm.*	3.5	3.5

* These readings were obtained on January 10, 1953.

one sample was 3.96 cc. taken in a sample from Ball Creek in June, 1953.

Only 27 of the 282 samples exceeded one cc. in total volume. The average number of bottom organisms per month ranged from a low of 19.2 in February to a high of 93.1 collected in September (Table II).

The standard deviation of the total numbers of organisms in these samples varied from 31 per cent to 86 per cent of the mean. This is somewhat higher than the 20 per cent to 50 per cent mentioned by Allen (1951) in his discussion of data published by a number of workers. It should be noted that in only four of the eighteen months sampled did the standard deviation exceed 60 per cent of the mean.

Needham and Usinger (1956) in a comprehensive study of a single riffle area conclude that quantitative routine sampling in streams to determine weights and numerical data is impractical. They state that "194 samples would be required to give significant figures on total weight of organisms, and 73 samples would be necessary to give significant figures on total numbers at a 95 percent level

Table II

Mean number and volume of bottom organisms collected from stations in Ball and Shope creeks, Coweeta Experimental Forest, October, 1952 to April, 1954.

Month	No. samples	Total no. organisms	Total volume in cc.	Mean no. per sq. ft.	Standard deviation	Mean vol. per sq. ft.
Nov. 1952	6	281	4.85	46.8	40.64	0.81
Dec.	18	370	6.80	20.6	13.12	0.38
Jan. 1953	18	430	7.65	23.9	13.07	0.43
Feb.	18	345	2.60	19.2	16.53	0.14
Mar.	12	426	5.25	35.5	16.90	0.44
Apr.	18	1,408	8.04	78.2	42.99	0.45
May	6	259	2.00	43.2	21.18	0.33
June	18	1,129	15.52	62.7	32.93	0.86
July	18	1,191	8.72	66.2	21.88	0.48
Aug.	18	953	6.19	52.9	27.01	0.34
Sept.	18	1,676	12.62	93.1	56.54	0.70
Oct.	18	1,013	9.28	68.7	36.32	0.52
Nov.	6	264	2.70	44.0	18.41	0.45
Dec.	18	457	8.77	25.4	14.31	0.49
Jan. 1954	18	383	5.22	21.3	9.88	0.29
Feb.	12	615	6.42	51.3	39.48	0.54
Mar.	18	775	4.77	54.1	24.7	0.27
Apr.	18	1,483	7.49	82.4	41.23	0.42

of confidence." These workers do not state what they mean by significant figures, since no comparisons were being made. We assume that the stated number of samples would be required to keep the standard deviation within a certain percentage of the mean. The excessive number of samples required to reduce the standard deviation, however, does not obviate the possibility of making valid quantitative statistical comparisons based on a reasonable number of samples.

Although the standard deviation of the numbers and mass of organisms in the stream bottom samples was high in the Coweeta studies, it was possible to establish significant differences between silted and nonsilted areas (Tebo, 1955) and between areas with forested and cleared stream banks (Hassler and Tebo, 1958). These comparisons were based on six samples per month in treated and control areas.

The peak in abundance of aquatic insects in these streams occurs during September (Fig. 2), and it is concurrent with an increase in the numbers of the very abundant caddis flies, beetles and two-winged flies (Figs. 11, 19, 34). A gradual decrease in the number of aquatic insects occurs during the fall months until the minimum is reached during December and January. This decrease occurs in all major orders represented in the samples.

After the January low, the numbers increased to a peak during April. The April high coincides with the peak of abundance of May flies and is attributed almost entirely to May flies and two-winged flies. The increase among two-winged flies, after the winter low, begins somewhat sooner than among the May flies, and the two-winged flies are at a fairly high level of abundance during February and March. There is a considerable decrease in all groups during May, and this decrease is associated with a major period of emergence of all groups except Trichoptera. After this May decrease in numbers, comparatively high levels are maintained during the summer months.

Peaks in the volume of aquatic insects occurred during November, 1952, June, 1953, and September, 1953. The November, 1952, and June, 1953, highs coincide with the maximum abundance of two large crane-fly larvae, *Tipula abdominalis* Say and *Tipula (A.) Williamsiana*. The high in September volume coincides with a peak in the number of organisms.

It is our opinion that numbers of aquatic organisms in the Coweeta samples give a better appraisal of the fauna than does volume. In a stream which has low standing crops of bottom fauna, large specimens, although scarce, give a distorted picture in terms of volume. While these large specimens may provide an occasional "bonus meal" and are utilized by larger trout, their scarcity makes it improbable

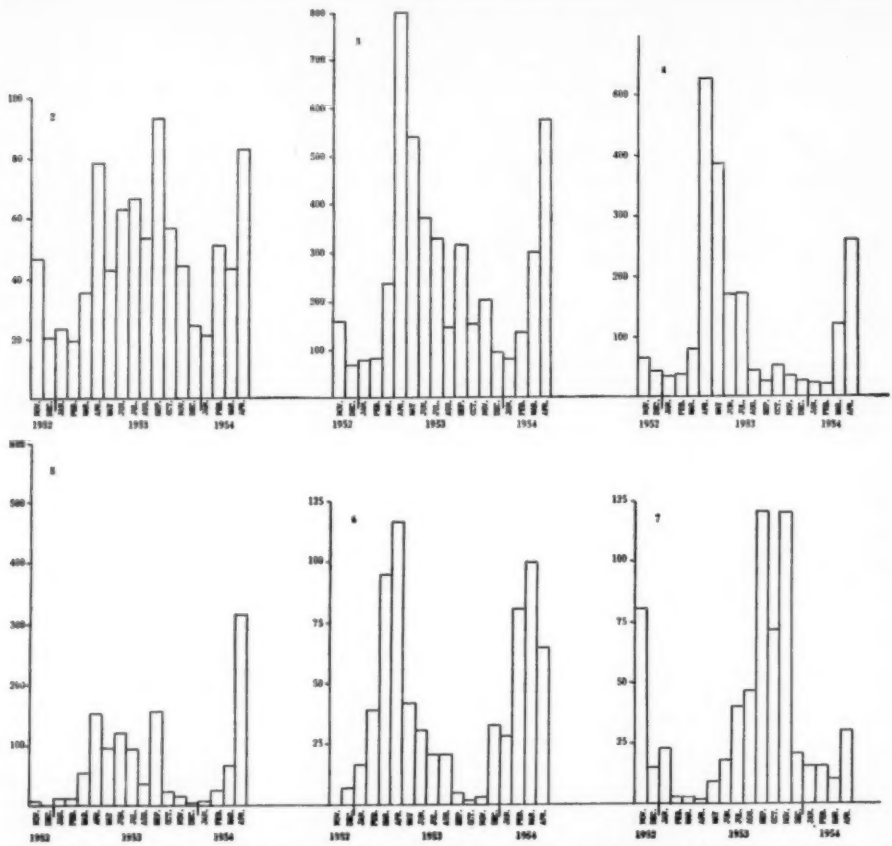


FIG. 2. Mean number of bottom organisms collected per month from Shope and Ball creeks, Coweeta Experimental Forest, from November, 1952, through April, 1954. FIGS. 3–7. Total number of nymphs collected per month from Shope and Ball creeks, Coweeta Experimental Forest. Adjusted to eighteen samples per month. Number of samples actually collected is given in Table III. FIG. 3. Ephemeroptera nymphs. FIG. 4. *Ephemerella spp.* FIG. 5. *Pseudocloeon spp.* FIG. 6. *Epeorus spp.* FIG. 7. *Stenonema sp.*

that they will be of importance in the daily foraging of the trout population.

The three orders—Ephemeroptera, Trichoptera, and Diptera, in that rank—comprised approximately three-fourths of the total collections for the eighteen-month period (Table III). The remaining one-fourth was composed of Plecoptera and Coleoptera. The Odonata and Oligochaetes contributed less than 1 per cent of all organisms collected. Except for the absence of the Megaloptera from the Coweeta collections, this distribution is similar to that found by Needham (1938), Surber (1951), and others. Although Megaloptera are present in Southern Appalachian streams, they are relatively scarce and were not collected in any of the Coweeta streams.

Representatives of the Coleoptera, comprising 10.5 per cent of the total collection, were more abundant in the Coweeta streams than in streams sampled in other parts of the country. The peak representation occurred during November and December, and this may account for their relative insignificance in studies carried out during the warmer months of the year (Table III). Aquatic Coleoptera were also abundant in the winter samples collected from Big Spring Creek in Virginia by Surber (1936).

There is a striking difference in the composition of the population by orders during different months of the year (Table III). The caddis flies are the dominant order from June through October. During November and

Table III

Total number and percentage composition of major groups of bottom organisms collected from Shope and Ball creeks, Coweeta Experimental Forest (per cent by months).

Order	Total no.	% total	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.
Ephemeroptera	4,657	30.4	18.5	17.5	17.7	23.6	41.2	57.2	69.0	32.7	27.5
Trichoptera	3,330	21.8	26.0	25.8	17.7	2.9	5.7	6.1	3.9	34.8	34.8
Diptera	3,279	21.4	16.0	16.0	25.1	60.0	36.9	22.1	15.5	10.7	9.8
Plecoptera	2,349	15.4	26.3	26.6	23.0	7.9	9.4	11.3	8.1	14.0	22.5
Coleoptera	1,602	10.5	12.8	12.9	16.0	5.0	6.9	3.2	3.5	7.1	4.8
Odonata	49	0.3	.0	0.8	0.5	0.3	.0	0.1	.0	0.6	0.5
Oligochaetes	28	0.2	0.4	0.3	.0	0.3	.0	.0	.0	.0	0.1

Order	Aug.	Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.
Ephemeroptera	15.3	18.7	15.4	25.4	20.0	19.8	17.9	38.6	32.6
Trichoptera	49.1	32.4	30.6	19.3	24.1	13.7	10.9	9.4	12.8
Diptera	9.9	22.7	12.7	9.5	12.9	34.5	44.3	34.3	28.8
Plecoptera	17.3	12.9	19.0	23.1	17.8	12.1	23.0	8.6	7.8
Coleoptera	7.0	13.2	21.4	22.7	19.7	13.3	14.6	8.4	7.3
Odonata	0.7	0.1	0.8	.0	0.2	0.5	0.1	0.8	0.1
Oligochaetes	0.7	0.1	0.0	0.0	.0	0.5	0.1	.0	0.7

December the stone flies gain ascendance by a very narrow margin. In January and February the larvae of two-winged flies are much more abundant than the other orders, and the fauna is completely dominated by May flies during March, April, and May.

Representative specimens of certain groups were sent to the following taxonomists for identification and verification of specimens already identified by project personnel: Ephemeroptera, E. W. Hamilton; Trichoptera, Dr. H. H. Ross; Plecoptera, Dr. A. R. Gaufin; Tipulidae, Dr. J. Speed Rogers; Coleoptera, Dr. Harry G. Nelson. Dr. H. K. Townes identified the specimens from approximately fifty of the samples included in this report.

Because of the lack of definite characters and because many of the nymphs and larvae have not been reared and associated with the adult, it is impossible to identify positively the nymphs and larvae of many aquatic insects to the species level. Many of those which have positive characters can be identified only from the mature nymph or larvae. In cases where early instar larvae and nymphs could not be separated, all specimens were grouped under the genus.

Ephemeroptera

May-fly nymphs, represented by twelve genera, were the most abundant order of insects in the total collections. The group reaches its peak of abundance during April, gradually decreases throughout the summer months, and attains a minor peak of abundance during September (Fig. 3). The seasonal low occurs during the winter months of December, January, and February.

With few exceptions, it was impossible positively to assign the May-fly nymphs to species.

Ephemerella spp. The genus Ephemerella, represented by at least eight species (including *E. bicolor, E. conestee, E. doris, E. dorothea, E. funeralis, E. fuscata, E. longicornis,* and *E. wayah*) is the most abundant genus of May flies in the collections Although the several species were grouped in Figure 4, the genus exhibits well-defined peaks and lows The peak occurs during April, and this peak, which is accompanied by the presence of the genus Epeorus, is primarily responsible for the April high for Ephemeroptera.

Pseudocloeon sp. and Baetis sp. These two genera were combined because of the impossibility of separating early instar nymphs. Most of the mature nymphs sorted belonged to the genus Pseudocloeon. These genera attain peaks in April and September and are relatively abundant during the summer months. They are least abundant from October through February (Fig. 5).

Epeorus spp. Epeorus, the third most abundant genus of May flies, is represented by the unidentified nymphs of several species. The

peak of abundance of the nymphs occurs during March and April, with well-defined lows during September, October, and November (Fig. 6).

Stenonema Spp. Nymphs of this genus, represented by *S. pudicium, S. carolina,* and *S. annexum* in the adult collections, are very abundant during September, October, and November and are at a very low level from December through June (Fig. 7).

Baetisca carolina. This species, although not abundant in the samples, exhibited a well-defined peak during November, after which none were collected in the samples until the following April (Fig. 8).

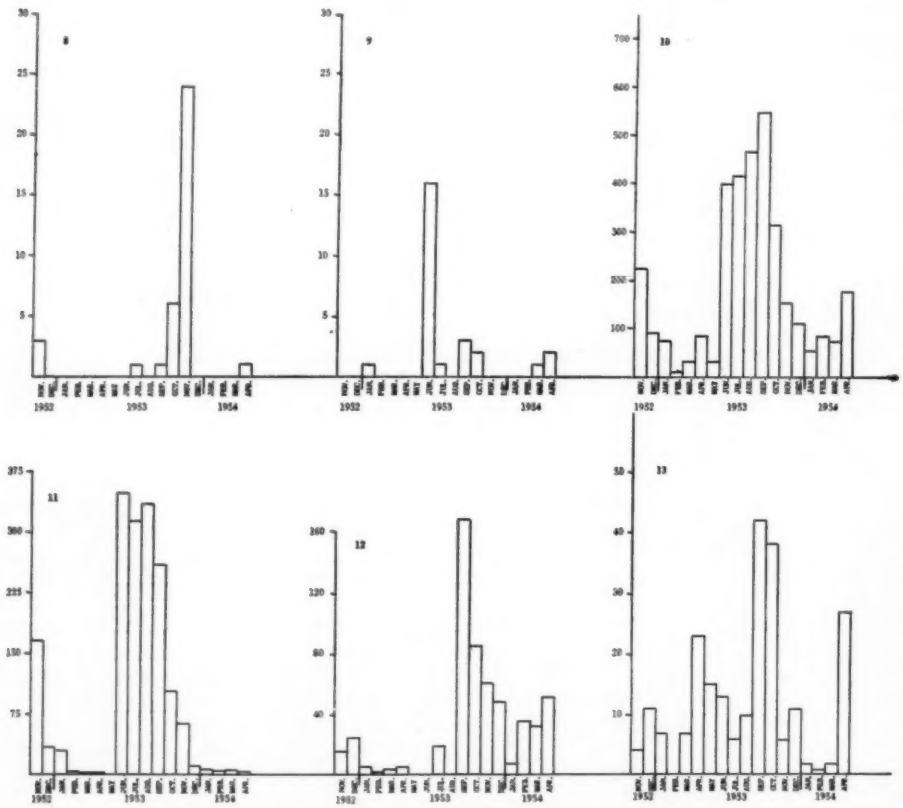
Isonychia sp. This genus, represented by several species (adults of *I. thalia, I. georgiae,* and *I. aurea* were collected), exhibited a peak during June. A few specimens were picked up sporadically during other months of the year (Fig. 9).

Trichoptera

Many larvae could not be assigned to definite species because of the lack of definite characters and the fact that relatively few adult Trichoptera from the Southern Appalachians have been associated with the larvae.

Caddis flies were the second most abundant order and represented 21.8 per cent of the total collections (Table III). This group was the most diverse order, and twenty different genera were identified from the collections.

Caddis-fly larvae build up to a high population during June and maintain this level until the peak is reached in September (Fig. 10). During this same period, the caddis flies are the dominant order in the combined



FIGS. 8–13. Total number of nymphs or larvae of aquatic insects collected per month from streams of Cowetta Experimental Forest. FIG. 8. *Baetisca carolina* FIG. 9. *Isonychia spp.* FIG. 10. Trichoptera larvae. FIG. 11. *Brachycentrus sp.* FIG. 12. *Cheumatopsyche spp.* FIG. 13. *Trentonius distinctus.*

collections (Table III). The order is at its lowest abundance during January, February, March, and April.

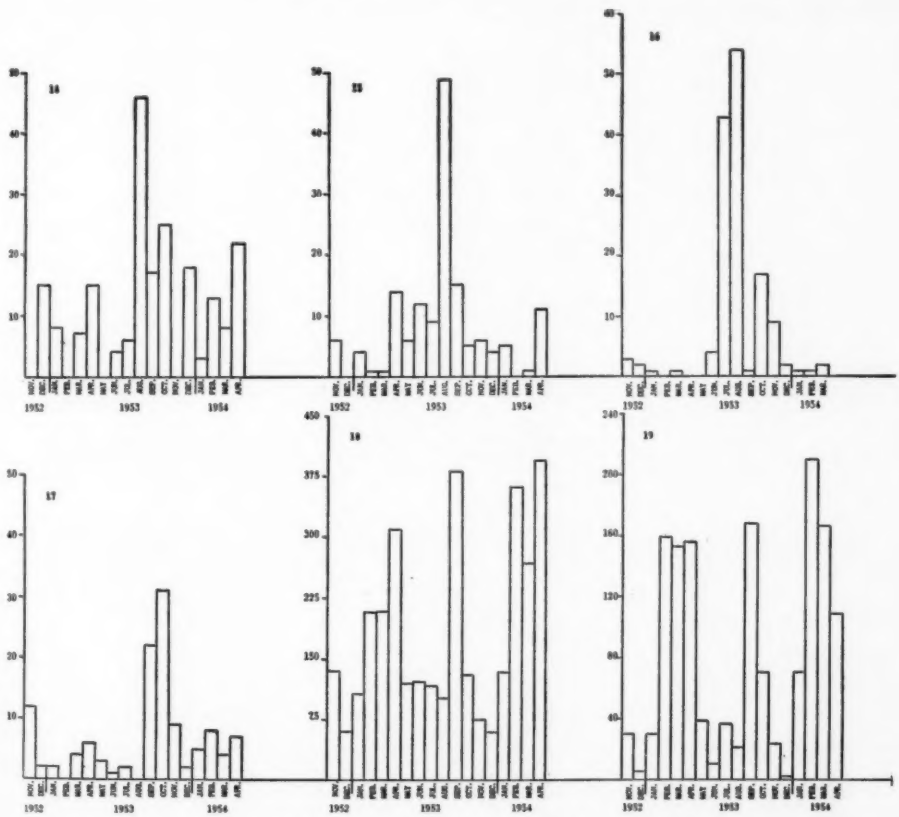
Brachycentrus sp. This is the most abundant caddis fly collected from the Coweeta Forest streams. Using Ross (1944), these specimens were all identified as *Brachycentrus numerous;* however no adults of this species were collected at Coweeta. The only adult member of the genus collected was *Brachycentrus spinae* Ross. The species exhibits a definite seasonal fluctuation and is most abundant in the stream during June through September, which coincides with the abundance for the whole order (Figs. 10 and 11). The species apparently emerges during early fall, and few larvae are collected from December through May.

Cheumatopsyche spp. This, the second most abundant genus of caddis flies, exhibits a definite peak during September, and after some fluctuation declines to a low during January. Few specimens are taken during the spring and summer (Fig. 12).

Trentonius distinctus. The larvae of this species exhibits two fairly well-defined peaks of abundance, one during April and another in September (Fig. 13). This corresponds with the reported production of adults during the entire year (Ross, 1944).

Diplonectra modesta. The larvae of this species fluctuates rather erratically, with a well-defined peak during August (Fig. 14). In other areas, the emergence is reportedly confined to spring and early summer (Ross, 1944). In the small, cold Coweeta streams, most of the adults appear to emerge during early fall.

Glossosoma nigrior. The larvae of this species also exhibits a well-defined peak during August



FIGS. 14–19. Total number of larvae of aquatic insects collected per month from streams of Coweeta Experimental Forest. FIG. 14. *Diplonectra modesta.* FIG. 15. *Glossosoma nigrior.* FIG. 16. *Hydropsyche sp.* FIG. 17. *Rhyacophila fuscula.* FIG. 18. Diptera. FIG. 19. *Simulium sp.*

but are not abundant in samples during the remainder of the year (Fig. 15).

Hydropsyche sp. Identifying larvae of this genus to the species level with certainty is difficult. However, the indication is that only one species (*Hydropsyche sparna*) was represented in the collections. Larvae of the genus exhibit a well-defined high during July and August, but are scarce during the remainder of the year (Fig. 16).

Rhyacophila fuscula. This very distinctive species was never abundant in the Coweeta streams. The larvae reached their peak in the stream samples during September and October (Fig. 17).

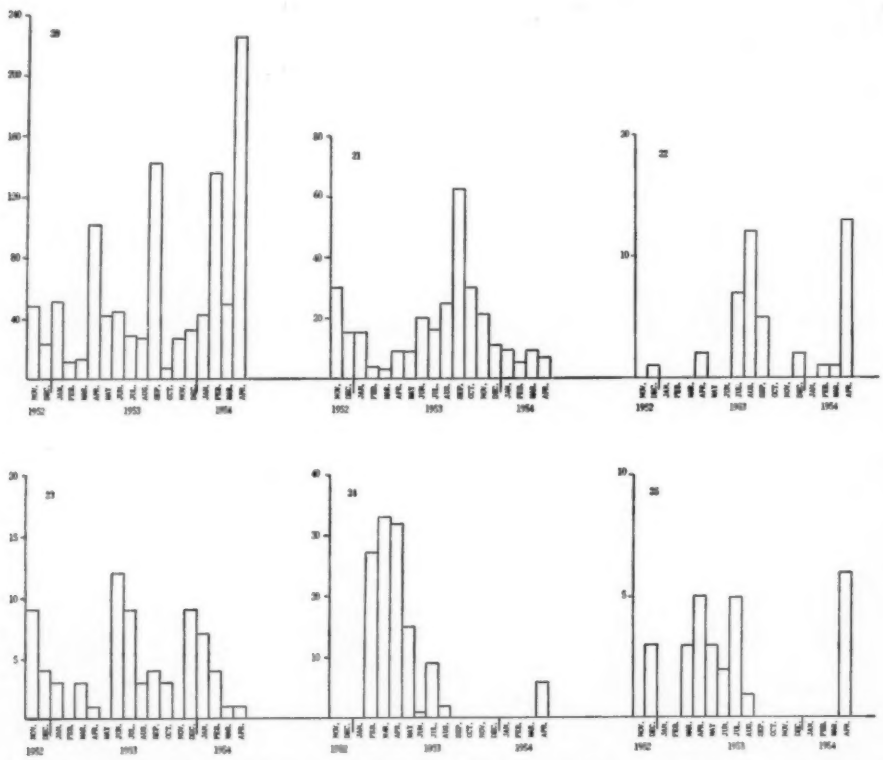
Diptera

The two-winged flies exhibit two major periods of abundance—in September and during the early spring months of February, March, and April (Fig. 18). This group becomes abundant earlier in the year than any of the other orders and is the dominant group in the collections during January, February, and March (Table III).

Simulium sp. Black-fly larvae were the most abundant two-winged flies collected in the samples. Their seasonal distribution is almost identical with that for the order (Figs. 18 and 19).

Tendipedidae. Members of the midge family, Tendipedidae, were the second most abundant group of two-winged flies. Not all of these specimens have been identified to date; however, the majority of midge larvae collected belong to the subfamily Hydrobaeninae. Again, the seasonal distribution of this group coincides closely with that for the order (Figs. 18 and 20).

Atherix variegata. This fly, a member of the snipe-fly family, Rhagionidae, was very abundant in the samples. The species is most



FIGS. 20–25. Total number of larvae of aquatic insects collected per month from streams of Coweeta Experimental Forest. FIG. 20. Tendipedidae FIG. 21. *Atherix variegata.* FIG. 22. *Blepharocera tenuipes.* FIG 23. *Tipula spp.* FIG. 24. *Antocha sp.* FIG. 25. *Protoplasa fitchii.*

abundant during September and the least abundant during February and March (Fig. 21).

Blepharocera tenuipes. This species reached its maximum abundance in the samples of February, March, and April of 1953 (Fig. 24). A few specimens were collected in the sampling from May through August and then the species was absent from the samples until April, 1954.

Tipula spp. This genus is represented by *Tipula abdominalis* and an unknown tipulid which may be *Tipula* (*a*). *williamsiana*, the adults of which were collected in the area. These two species were the largest two-winged fly larvae in the collections. Seasonal distribution was erratic with highs during November, 1952, June, 1953, and December, 1953 (Fig. 23).

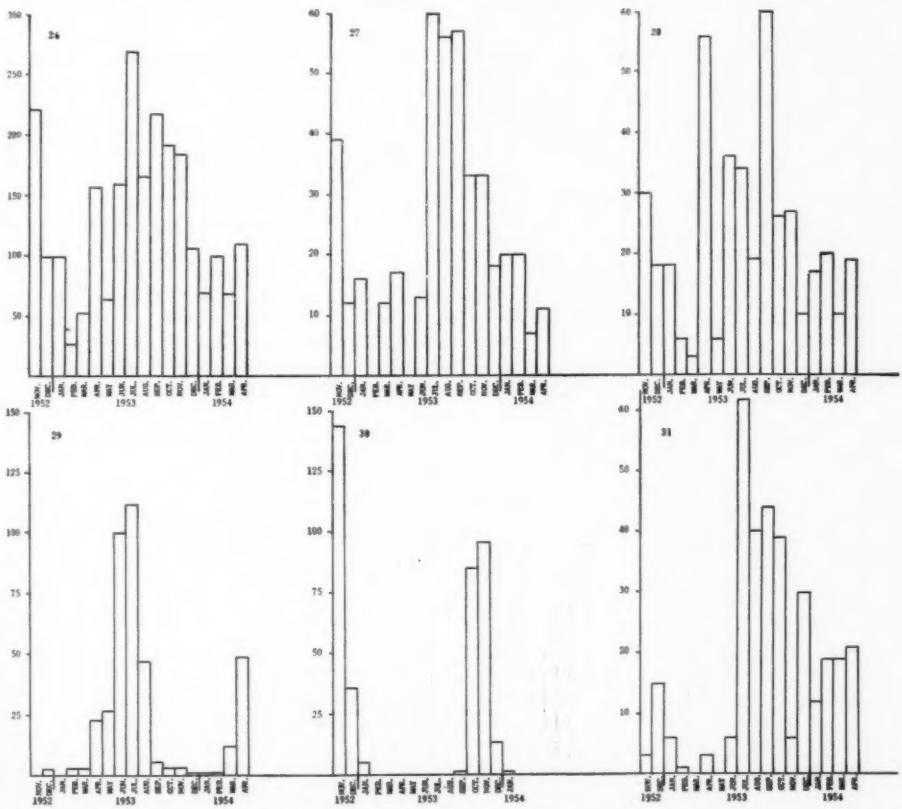
Antocha sp. This genus was not abundant in the samples and the largest number of specimens were collected during July, August, and April (Fig. 22).

Protoplasa fitchii. The larvae of this species were present in small numbers from March through August (Fig. 25).

Plecoptera

Stone flies ranked fourth in abundance among the orders of aquatic insects present in the samples (Table III). During November and December they were the dominant order in the total collections. The group reaches its maximum numbers in the samples during July and was at a high level from May through November (Fig. 26).

Peltoperla maria. This very distinctive stonefly nymph was the dominant species in the order. The species reaches its greatest abundance in the samples during July, August, and



FIGS. 26–31. Total number of nymphs of aquatic insects collected per month from streams of Coweeta Experimental Forest. FIG. 26. Plecoptera. FIG. 27. *Peltoperla maria.* FIG. 28. *Pteronarcys scotti.* FIG. 29. *Leuctra sp.* FIG. 30. *Taeniopteryx maura.* FIG. 31. *Acroneuria abnormis.*

September and is at a low level from December through June (Fig. 27).

Pteronarcys scotti. This stone fly was almost as abundant as *P. maria* and is the largest aquatic insect collected from Coweeta streams. Fully developed specimens reach a length of almost two inches. *P. scotti* nymphs require more than one year for development and the seasonal distribution is erratic, with highs during April and September (Fig. 28).

Leuctra sp. Some nymphs of this genus can probably be assigned to *Leuctra sara,* the adults of which were collected during February and April. The nymphs are very abundant in Coweeta streams with well-defined peaks of abundance during June and July (Fig. 29). Few specimens were collected during the fall and winter.

Taeniopteryx maura. The nymphs of another abundant stone fly, *T. maura,* reach their peak of abundance during October and November and are completely absent from the samples from February through August (Fig. 30). This species apparently emerges in midwinter and the adults are abundant during January.

Acroneuria abnormis. This stone-fly nymph is exceeded in size only by *P. scotti.* The nymphs are abundant in the streams from July through October with the peak in July (Fig. 31).

Brachyptera fasciata. The nymphs of this stone fly, similar in appearance to *T. maura,* occur somewhat later than *T. maura* and are abundant during December and January (Fig. 32). The nymphs are completely absent from most of samples from February through November.

A number of other stone flies were collected in the samples but were not present in sufficient numbers to measure their seasonal distribution.

Coleoptera

Because the adults and larvae of aquatic beetles were not recorded separately in some samples, it is not possible to compare the seasonal distribution of the larvae.

The largest collections of beetles occurred during the months of September, October, and November (Fig. 33). The beetle fauna was composed almost entirely of two species—*Anchytarsus bicolor* and *Heterlimnius ovalis.*

Odonata

The only dragonfly nymph collected from Coweeta streams was a member of the genus Lanthus. This nymph was not abundant and comprised only 0.3 per cent of the entire collections (Table III).

Summary

A total of 267 square-foot samples were collected over an eighteen-month period at three sampling stations on Shope and Ball Creeks, Coweeta Experimental Forest, Macon County, N. C. These streams had low standing crops of bottom fauna and the monthly average ranged from a low of 19.2 organisms per square foot during February to a high of 93.1 organisms per square foot during September.

The average monthly volume ranged from a low of 0.14 cc. during February to a high of 0.86 cc. during June. There were two peaks in the numerical abundance of the bottom organisms with the major peak in September and

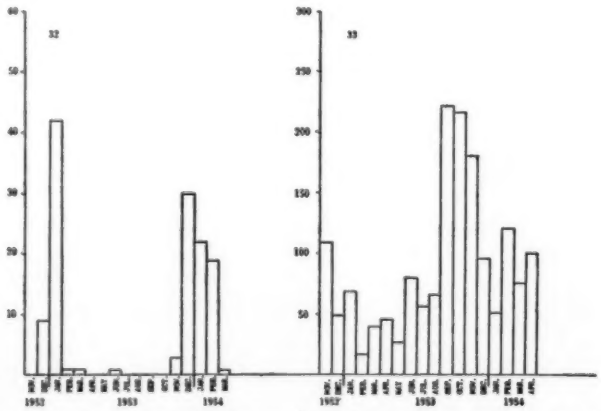


FIG. 32. Total number of nymphs of *Brachyptera fasciata* collected per month. FIG. 33. Total number of Coleoptera larvae and adults collected per month.

a lesser peak during April. Bottom organisms were least abundant during December and January in Shope and Ball creeks.

The three orders Ephemeroptera, Trichoptera, and Diptera—in that rank—comprised approximately three-fourths of the total collection. The remaining one-fourth was composed of Plecoptera and Coleoptera. Odonata and Oligochaetes comprised less than 1 per cent of all organisms collected.

There was considerable variation in the species composition during different months of the year. Trichoptera were dominant from June through October, Plecoptera during November and December, Diptera during January and February, and Ephemeroptera from March through May.

LITERATURE CITED

ALLEN, K. RADWAY. 1951. The Horokiwi Stream. New Zealand Marine Dept. Fish. Bull. No. 10.

HASSLER, W. W., AND L. B. TEBO, JR. 1958. Fish Management Investigations on Trout Streams. F. A. Proj. Comp. Report. Fish Division, N. C. Wildlife Res. Comm., Raleigh, N. C.

NEEDHAM, PAUL R. 1938. Trout Streams. Comstock Publishing Co., Ithaca, N. Y.

NEEDHAM, PAUL R., AND ROBERT L. USINGER. 1956. Variability in the macrofauna of a single riffle in Prosser Creek, California, as indicated by the Surber sampler. Hilgardia **24**: 383–409.

ROSS, HERBERT H. 1944. Caddis Flies, or Trichoptera, of Illinois. Bull. Ill. Nat. Hist. Surv. 23. 326 pp.

SURBER, E. W. 1936. Rainbow trout and bottom fauna production in one mile of stream. Trans. Am. Fish. Soc. **66**: 193–202.

———. 1951. Bottom fauna and temperature conditions in relation to trout management in St. Mary's River, Augusta County, Virginia. Va. Jour. Sci. **2**: 190–202.

TEBO, L. B., JR. 1955. Effects of siltation, resulting from improper-logging, on the bottom fauna of a small trout stream in the Southern Appalachians. Prog. Fish. Cult. **17**: 64–70.

BRYOPHYTES OF THE PRIMEVAL FOREST

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The vast primeval forest of the Blue Ridge, extending from northern Alabama and Georgia to New York, has almost disappeared. It seems important to record in as much detail as possible the composition of that forest before all traces of it are lost.

A remnant of the primeval hemlock-hardwood forest, about 30 acres, still survives 4½ miles east of Highlands, N. C., on U. S. Highway 64. The tract is part of the original Ravenel Forest, and now belongs to Mr. Henry Wright and his brother. It is part of the area studied by Oosting & Billings (1939). These authors have adequately described the vascular vegetation of the forest, thus giving an ideal foundation for the study of the bryophytes in relation to the communities of higher plants.

The area studied by Oosting and Billings was "roughly 1½ mile by 1 mile in extent," not burned, grazed, or cut. "From nearby elevations, the general aspect of the forest is that of a continuous hemlock stand."

The forest lies at about 35° 2′ N; elevation 4,400 feet. Temperatures at Highlands are mild, the January and July means being 39.7°F. and 70.4°F. respectively. The average precipitation is 79.77 inches, quite evenly distributed throughout the year. By months the averages are:

Jan.	7.37 inches	July	9.38 inches
Feb.	6.25	Aug.	7.98
Mar.	8.73	Sept.	5.33
Apr.	5.90	Oct.	4.86
May	5.11	Nov.	5.43
June	6.35	Dec.	7.58

Frequency and duration of droughts are important for plants, especially for epiphytic plants (Barkman, 1958). Van Bavel and Verlinden (1956) state that "in the mountains [of North Carolina] drought is non-existent in April, almost so in May, and rises to an expectancy of about 15 per cent in June and then becomes extinct again for the remainder of the season [April through September]."

It is interesting and sometimes useful to try to correlate the plant communities with environmental conditions (as shown by climatic and edaphic data) and historical relations both short term and long. But I have long believed that the plant community gives a better summation of all of the factors than the most precise instruments or the most competent

electronic computer. The plants are integrating all of the factors continuously, time without end. "Natural vegetation is still the best integrator of both [climate and soils] that we have" (Sears, 1960).

Oosting and Billings recognized two types of forest: a Rhododendro-Tsugetum (*Rhododendron* type) with undergrowth of *Rhododendron maximum* and *Kalmia latifolia*, and (2) a Vaccinio-Tsugetum (*Polycodium* type) with undergrowth of *Vaccinium stamineum*.[1] The former occupies three-fourth of the remaining virgin forest, much of it being an impenetrable thicket eight or ten feet high. Beside hemlocks (*Tsuga canadensis*) two to five feet in diameter and up to 450 years old, Oosting and Billings noted *Acer rubrum, Betula lenta, Castanea dentata, Halesia carolina, Liriodendron tulipifera, Magnolia fraseri*, and *Prunus serotina*. At present *Castanea* is gone, and we did not notice any *Liriodendron*.

The Vaccinio-Tsugetum had as "dominants" *Betula lenta, Castanea dentata, Quercus borealis* (including its var. *maxima*), and *Tsuga canadensis*.

Oosting and Billings devoted most of their attention to woody plants. One table names herbaceous plants—two species in the *Rhododendron* type, plus "mosses and liverworts" with frequency of 70 and percentage of cover of 7.5. This is the only reference to bryophytes. In the *Vaccinium* (*"Polycodium"*) type there were sixteen herbaceous species, with a total coverage of 57 per cent. The soils of both types were extremely acid: pH 3.06 to 3.63. They were also extremely light and loose, especially in the *Rhododendron* type, where organic matter makes up "95.51% on a dry weight basis."

Apparently the Vaccinio-Tsugetum occupies drier sites. Various shrubs and young trees occur in both communities, as well as fallen logs in all stages of decay. We found the ground cover in both types to be deep, loose duff (Oosting and Billings' fine mull), with no vegetation except an occasional *Chimaphila maculata, Goodyera repens*, or *Mitchella repens*.

The Rhododendro-Tsugetum

Fallen logs are covered with bryophytes. Tree and shrub trunks are more or less covered. The giant hemlocks rarely have any bryophytes above six feet from the ground, though they are often completely covered on the lowest two or three feet. Extensive mats on bases of trees are composed of *Thuidium delicatulum, Bazzania trilobata, Hypnum imponens, H. reptile*, and *Brotherella recurvans*, with the last predominating. Extensive mats of the same species occur on fallen logs. On half-buried logs *Dicranum scoparium* forms large cushions. Occasional pads of *Leucobryum glaucum* (sterile) occur. In some cases *Nowellia curvifolia* covers a rotting log.

On eleven living hemlocks we found fifteen species of bryophytes. The most abundant species were *Brotherella recurvans* on eight trees, *Dicranum montanum* on six, *D. fulvum* on eight, *Hypnum reptile* on nine, *Heterophyllium nemorosum* on four, *Paraleucobryum longifolium* on three, and *Tetraphis pellucida* on three (at base of trees only).

On maple, living and fallen, 34 species were found. A recently fallen (1958) 18-inch maple gave opportunity to study bryophytes up to 54 feet above the ground. Most abundant were: *Anomodon attenuatus*, 0–24 feet; *Dicranum fulvum*, 6–13 feet; *Fissidens subbasilaris* (coming in after the tree fell); *Leucodon brachypus*, 6–34 feet. *Frullania brittoniae*, 6–30 feet; *Platygyrium repens*, 3–54 feet; *Porella platyphylloidea*, 3–33 feet; *Schwetschkeopsis denticulata*, 3–24 feet; *Ulota crispa*, 15–54 feet; *Metzgeria crassipilis*, 27–54 feet (three collections); *Microlejeunea ulicina*, 0–30 feet.

On *Rhododendron maximum* (trunks up to eight inches in diameter) we found *Bazzania trilobata, Brotherella recurvans, Heterophyllium nemorosum, Hypnum reptile, Microlejeunea ulicina, Plagiothecium denticulatum*, and *Radula obconica*.

On *Kalmia latifolia* (large, old trunks) we found *Bazzania trilobata, Brotherella recurvans, Hypnum reptile, Metzgeria furcata, Microlejeunea ulicina, Plagiothecium denticulatum*, and *Radula obconica*.

On a rotten log were found *Nowellia curvifolia* and *Odontoschisma denudatum*.

The fitness of the habitat is indicated by the occurrence of capsules. These were observed on *Anacamptodon splachnoides, Brotherella recurvans* (abundantly), *Dicranella heteromalla, Dicranum flagellare, D. fulvum, D. scoparium, Funaria hygrometrica* (on a disturbed area), *Heterophyllium nemorosum, Homalotheciella subcapillata, Hypnum imponens, H. reptile* (abundantly), *Tetraphis pellucida, Ulota ludwigii*, and *U. crispa*.

A small stream crosses the tract from south

[1] Latin names of vascular plants herein are from Gray's Manual (Fernald, 1950); of Hepatics from Frye and Clark (1937–1947) or Schuster; of Mosses from Grout (1928–1937); *Sphagnum* from Andrews (1913).

to north. In the vicinity of the stream bryophytes are especially abundant. On wet rocks and sandy banks several species occur which are not found elsewhere: *Atrichum crispum, A. undulatum* (very large), *Diphyscium cumberlandianum, Hookeria acutifolia* with gemmae at the tips and on surfaces of leaves, *Mnium affine, M. punctatum* (small rock form and large land form), *Pellia* sp., *Plagiothecium elegans* with axillary and terminal gemmae, *Scapania nemorosa, Sphagnum palustre.*

Bryophytes of the Rhododendro-Tsugetum with the Number of Records of Each

Amblystegium juratzkanum 1
serpens 1
varium 1
Anacamptodon splachnoides 1
Anastrophyllum michauxii 1
Anomodon attenuatus 14
rostratus 2
rugelii 1
tristis 5
Atrichum angustatum 1
crispum 1
undulatum 1
Bazzania trilobata 20
Blepharostoma trichophyllum 1
Brachythecium oxycladon 3
salebrosum 2
Brotherella recurvans 36[2]
Bryum argenteum 1 (on a disturbed area)
Calypogeia fissa 2
neesiana 1
Campylium chrysophyllum 2
hispidulum 1
Cephalozia catenulata 2
media 5
Dicranella heteromalla 1
Dicranum flagellare 3
fulvum[3] 30
montanum 7
scoparium 3
Diphyscium cumberlandianum 1
Diplophyllum apiculatum 1
Entodon brevisetus 2
Eurhynchium serrulatum 1
Fissidens subbasilaris 8
Fontinalis dalecarlica (on abutment of bridge at roadside)
Frullania asagrayana 13
brittoniae 7
eboracensis 4
Funaria hygrometrica (on a disturbed area)
Harpalejeunea ovata 7
Heterophyllium nemorosum 16
Homalotheciella subcapillata 12
Homomallium adnatum 1
Hookeria acutifolia 2
Hylocomium brevirostre 1 (on base of tree)
Hypnum imponens 12
fertile 1
reptile 33
Jubula pennsylvanica 1
Lejeunea lamacerina 2
Leptodon trichomitrion 1
Leucobryum glaucum 3
Leptodictyum trichopodium 1
Leucodon brachypus 23
Lophocolea cuspidata 1
Metzgeria crassipilis 5
fruticulosa 3
furcata 9
Microlejeunea laetevirens 1
ruthii 1
ulicina 19
bullata 1
an intermediate form 1
Microlepidozia sylvatica 2
Mnium cuspidatum 1
punctatum 2
Neckera pennata 9
Nowellia curvifolia 12
Odontoschisma denudatum 1

[2] A very slender form of *Brotherella recurvans* is common on trees in the primeval forest. I believe it is a form modified by the flow of water along the trunk in times of rain. On one dead and exposed tree down by the brook, *Heterophyllium nemorosum* covers several square feet, dangling in slender, loose, small-leaved strands, presumably another water-flow form. *Hypnum cupressiforme* var. *filiforme* in western Europe is another, being, according to Barkman, "restricted to dry, smooth, hard, vertical, acid substrates [rocks and bark] in shady places." I have previously noted that the western North American *Homalothecium nuttallii* var. *hamatidens* is only a pendulous form of the species, often found along the pendant margin of a sod of the species. Probably *Isothecium myosuroides* var. *filiforme* is another water-flow form. The water current is responsible for combing out the strands into parallel skeins. The slenderness of the shoots may be due to the dilution of the nutrient solution available to the plant, resulting in mineral starvation.

[3] This *Dicranum* on the bark of trees has all of the morphological characteristics of *D. fulvum*, which normally grows on rocks, and does so in the cleared area to be mentioned later. Being on a tree, it should be *D. viride*, but the leaves are not broken, as is characteristic for *D. viride*. I yield to the advice of Dr. L. E. Anderson in calling it *D. fulvum*. Dr. Anderson states that true *D. viride* also occurs in North Carolina. Could the broken leaves be due to xerophily?

Table I

10-meter quadrat in Vaccinio-Tsugetum. August 11, 1959. Primeval Forest, 4½ miles east of Highlands, N. C., on U.S. Highway 64. Ground nearly level, covered with duff and fallen leaves; no soil exposed.

	Cover, %	Vitality*	Periodicity†	
CANOPY				50 TO 70 FT. ABOVE GROUND
Tsuga canadensis	33	1	fol.	2½ ft. diam.; 14 in. diam.
Acer rubrum	25	1	fol.	15 in. diam.
Fraxinus americana	5	1	fol.	15 in. diam.
Magnolia fraseri	10	2	fol.	10 in. diam.
TALL SHRUB LAYER				15 FEET TALL
Hamamelis virginiana	15	3	fol.	
Clethra alnifolia		4	fol.	
Betula lenta		3	fol.	
Oxydendron arboreum	1	1	fl.	
LOW SHRUB LAYER				1 TO 3 FEET TALL
Vaccinium stamineum	50	2	fol.	
GROUND LAYER				
Dryopteris noveboracensis		3	fol.	
Uvularia sessilifolia		4		70 plants
Quercus rubra sdl.			fol.	
Chimaphila maculata		4	fol.	
Acer rubrum sdl.			fol.	
Sassafras albidum			fol.	
Trillium sp.		2	fol.	
Mitchella repens		3	fol.	
Goodyera pubescens		1	fl.	
Smilax rotundifolia		3	fol.	
BRYOPHYTES				
Bazzania trilobata		2		On half-buried rotten log
Dicranum scoparium		1	caps.	" " " " "
Heterophyllium nemorosum		2		" " " " "
Hypnum imponens		1		" " " " "
Leucobryum glaucum		3		" " " " "
Brotherella recurvans				On Hamamelis
Frullania asagrayana				" "
Hypnum reptile				" "
Leucodon brachypus				" "
Radula caloosiensis				" "
Schwetschkeopsis denticulata				" "
Thuidium delicatulum				" "
Dicranum fulvum				" maple
Leucodon brachypus				" " , 6 ft. up
Thuidium delicatulum				" "
Brotherella recurvans		2		On a tree
Dicranum montanum		2		" " "
fulvum		2		" " "
Frullania asagrayana		1		" " "
brittoniae				" " "
Harpalejeunea ovata		3		" " "
Herberta tenuis		2		" " " , outside quadrat
Homalotheciella subcapillata		1		" " "
Hypnum reptile				" " "
Metzgeria fruticulosa		4		" " "
furcata		4		" " "

Microlejeunea ulicina		2		" " "
Radula andicola		3		" " "
Schwetschkeopsis denticulata		3		" " "
Cephalozia media				On small decorticated logs on the ground
Nowellia curvifolia				" " " " " " "
Odontoschisma denudatum		2		" " " " " " "
Tetraphis pellucida		1		" " " " " " "

* Vitality: 1 = Regularly completing the life cycle; 2 = Strong, but usually not completing the life cycle; 3 = Feeble, but spreading; 4 = Occasionally surviving, but not increasing.
† Periodicity: Fol. = in leaf; Fl. = in flower.

Paraleucobryum longifolium 8 (small tufts on trees, not falcate).
Pallavicinia lyellii 1
Pellia sp. 1
Plagiochila caduciloba 2 (on trees!)
Plagiothecium denticulatum 2
elegans 8
Platygyrium repens 12
Polytrichum ohioense 1
Porella platyphylloidea 8
Radula andicola 1
caloosiensis 14
obconica 2
tenax 1
Rhabdoweisia denticulata 1
Rhodobryum roseum 3
Scapania nemorosa 1
Sciaromium lescurii 1
Schwetschkeopsis denticulata 16
Riccardia palmata 1
Sematophyllum carolinianum 1
Solenostoma crenulata gracillima 1
Sphagnum palustre 2
Tetraphis pellucida 6
Thuidium delicatulum 6
minutulum 3
Trichostomum cylindricum 2
Tritomaria exsecta 1
Ulota crispa 8
Zygodon viridissimus 1

The numbers in the above list are only suggestive. The most abundant species were seen by all of us many more times than the records indicate. On the other hand the fallen maple was studied in three-foot lengths, and the bryophytes were recorded for each three feet. Thus the records of *Fissidens subbasilaris, Leucodon brachypus, Platygyrium repens,* and *Schwetschkeopsis denticulata* were nearly all from that one tree. *Sphagnum palustre* was in one big swampy area where the brook approaches the highway.

The Vaccinio-Tsugetum

The Vaccinio-Tsugetum, of which only a small sample remains undisturbed, was analysed by means of a ten-meter quadrat. The record appears in Table I.

Cut-Over Land

Adjacent areas of the Ravenel Forest were lumbered in the 1940's. A few old maples and hemlocks were left standing. In a former Vaccinio-Tsugetum there is an open stand of *Vaccinium stamineum,* with a low, loose canopy of oak saplings (*Q. alba, rubra, prinus*). The ground is covered with oak leaves, without any ground vegetation. There is no reproduction of maple, birch, magnolia, or hemlock. In this community, across an abandoned roadway from the Rhododendro-Tsugetum, the following bryophytes were seen:

Anomodon attenuatus
tristis
Brachythecium acutum
Brotherella recurvans
Campylium chrysophyllum
Dicranum flagellare
fulvum (on rocks and on trees)
scoparium
Fissidens subbasilaris
Frullania asagrayana
brittoniae
Hedwigia ciliata (on rocks)
Heterophyllium nemorosum
Hylocomium brevirostre
Hypnum imponens
reptile
Leptodon trichomitrion
Leucobryum glaucum
Leucodon brachypus
Mnium affine
Platygyrium repens
Schwetschkeopsis denticulata
Thuidium delicatulum
minutulum
Ulota americana
crispa
ludwigii

This stand may be compared with the virgin stand of the same community: Number of species

in virgin stand, 26; in cleared land, 30. Significant species lacking on cleared land: *Herberta tenuis, Metzgeria furcata, Odontoschisma denudatum, Paraleucobryum longifolium, Riccardia palmata.*

These three types of forest show the effects of exposure to xeric conditions. The tabulation indicates the fitness of the environment for the mosses. The absence of sporophytes in mosses may not indicate any lack of vegetative fitness. Vast areas of luxuriant mosses occur without ever producing antheridia or archegonia (acres of *Rhytidium rugosum* in northern Ireland and square rods of it in northeastern Iowa; *Brachythecium oxycladon* in Iowa; *Thuidium delicatulum* in this tract). The presence of *so many* sporophytes in the primeval forest means something about environmental conditions. This meaning can only be deduced by a comparison with similar studies of other stands and other communities.

Billings and Drew (1938) made a special study of corticolous bryophyte communities in a Liriodendro-Tsugetum on the west side of the Great Smoky Mountains at "about 3200 ft." elevation. This community is radically different from ours at Highlands, but there are several species common to both. Two trees of *Tsuga* and one of *Castanea dentata* were examined by Billings and Drew. They found that "unions" of *Ulota crispa, Dicranum fulvum-Bazzania denudata, Hypnum reptile, Bazzania trilobata,* and *Mnium hornum* were exclusive to the bark of *Tsuga.* (The bark of *Tsuga* is the driest of those studied, and the most acid *p*H 4.28–4.96).

The presence of mosses (*Ulota*) in abundance in the crowns of hemlocks in the Liriodendro-Tsugetum of the Smokies indicates a wetter climate than that of Highlands. Doubtless the clouds that so constantly shroud the Smokies, and give them their name, keep the tree tops well watered. We noticed at Highlands that hemlocks by the brook carried mosses higher up than those on the terraces above the valley of the brook. This small valley is certainly more humid than the terraces. Beside, in our area the "canopy" is far below the tops of the mostly isolated, and therefore fully exposed, hemlocks.

Patterson (1940) at Mountain Lake, Va., recognized "9 well developed bryophyte communities." One area studied had a *"Tsuga Canadensis-Rhododendron* coverage." Patterson found the bark of *Tsuga* to be much drier than any other bark and much more acid (*p*H 4.3).

Comparing our records with those of Billings and Drew and of Patterson, it is notable that where we had *Betula lenta,* Billings and Drew had *B. lutea* and Patterson had *B. lenta. Magnolia* is not mentioned in either of these two papers. And *Fagus,* present in Billings and Drew and Patterson, was absent at our site. *Fissidens subbasilaris* of our records and of Patterson is absent from Billings and Drew; its place is taken there by *F. cristatus,* growing in mats at bases of trees. *Ulota crispa* was abundant in crowns of hemlocks in Billings and Drew, but we found it only on deciduous trees.

In all of these three areas *Ulota crispa, Neckera pennata, Dicranum fulvum* (on trees), and *Microlejeunea ulicina* were found. At Highlands only we found *Radula caloosiensis, R. tenax, Entodon brevisetus, Heterophyllium nemorosum, Herberta tenuis, Diphyscium cumberlandianum, Harpalejeunea ovata, Zygodon viridissimus, Hookeria acutifolia, Jubula pennsylvanica,* and others, revealing a wholly unique community. But the large number of widespread species at Highlands permits comparison with hardwood-hemlock forests elsewhere. Where our plant communities have been minutely studied we shall be using them, rather than the figures from batteries of instruments, to define the soil-climate complex.

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LITERATURE CITED

ANDREWS, A. LEROY. 1913. Sphagnaceae in North American Flora. New York Botanical Garden.

BARKMAN, J. J. 1958. Phytosociology and Ecology of Cryptogamic Epiphytes. Van Gorcum & Co.: Assen, Netherlands.

BILLINGS, W. D., AND W. B. DREW. 1938. Bark factors affecting the distribution of corticolous bryophytic communities. Amer. Midl. Nat. **20** (2): 302–330.

CARNEY, C. B. 1958. Weather and Climate in North Carolina. Bull. 396 reprint, Agric. Exp. Sta., North Carolina State College, Raleigh, N. C.

CONARD, H. S. 1951. *Homalothecium nuttallii*. The Bryologist **54**: 74–75.

FERNALD, M. L. 1950. Gray's Manual of Botany. Amer. Book Co.

FRYE, T. C., AND LOIS CLARK. 1937–1947. Hepaticae of North America. University of Washington Press: Seattle, Wash.

GROUT, A. J. 1928–1937. Moss Flora of North America. Publ. by the author, Newfane, Vt.

OOSTING, H. J., AND W. D. BILLINGS. 1939. Edapho-vegetational relations in Ravenel's Woods. Amer. Midl. Nat. **22** (2): 333–350.

PATTERSON, P. M. 1940. Corticolous bryophyte societies at Mountain Lake, Virginia. Amer. Midl. Nat. **23** (2): 421 441.

SEARS, PAUL B. 1960. The place of ecology in science. Amer. Nat. **94**: 193–200.

VAN BAVEL, C. H. M., AND F. J. VERLINDEN. 1956. Agricultural Drought in North Carolina. Tech. Bull. No. 122. North Carolina Agric. Exp. Sta., Raleigh, N. C.

A DISPOSITION OF THE NOMENCLATURE OF LICHENS IN CURTIS'S NORTH CAROLINA *BOTANY* OF 1867

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In preparing the lichenological works of the American botanist Edward Tuckerman (1817–1886) for republication (Culberson, 1960), I excluded from the book one paper that had been attributed to him. It is the treatment of the lichens in M. A. Curtis's (1867) *Botany* of North Carolina. Although there are good reasons to exclude this catalogue from such a collection, it contains some unlisted names that must be dealt with.

Tuckerman prepared his list of lichens for Curtis in 1860, but the outbreak of the Civil War prevented its publication. After the war Tuckerman (1866) wrote to Curtis that "there are very important reasons for the revision of the N[orth] Carolina Catalogue of Lichens. As it is, it will be far from expressing my matured views." In the next year Curtis's *Botany* was published, the lichenological section—listing 218 species—being attributed to Tuckerman in the preface. In the publication this list was much modified from Tuckerman's holographic manuscript of 1860 (now preserved among the Moses Ashley Curtis Papers in the library of the University of North Carolina, Chapel Hill). The changes were mainly Curtis's deletion of Tuckerman's annotations and remarks for species and the unfortunate elimination of the localities for specimens which left the impression that all the records were for North Carolina while in reality many were for South Carolina and Virginia. Tuckerman was annoyed at this abuse of his work, and Henry Willey (1886) wrote of this paper that Tuckerman "refused to acknowledge it as it had been made up from an old list with changes and additions which he had not been permitted to see."

Curtis's obscure publication in Raleigh went unnoticed by contemporary European lichenologists and Tuckerman, understandably, did not call it to their attention. Later none of the lichen names in it were recorded in Zahlbruckner's monumental ten-volume *Catalogus Lichenum Universalis* (1922–1940).

The unlisted names in the North Carolina catalogue fall into the three categories below. The new combinations and names are attributed to their authors (for the most part, Tuckerman) "ex Curtis." All but one of these unlisted names appear in Tuckerman's manuscript, and his later objections to the published version seem unrelated to the present problem of assigning authority. Some lichenologists, however, will prefer to attribute all these names to Curtis alone, and such practice is quite permissible.

Of Tuckerman's five new combinations, *Coccocarpia parmelioides* and *Lecanora myrini* are especially important now because they involve a change in the previously accepted authority of the current names of two species which are widely distributed and locally common here and abroad.

I wish to thank Dr. I. Mackenzie Lamb (Harvard University) and Dr. R. L. Wilbur (Duke University) for advice on points of nomenclature and Dr. James Patton of the Southern Historical Collections, University of North Carolina Library, for permission to examine Tuckerman's manuscript on the lichens and his letters to Curtis.

1. New Combinations

Coccocarpia parmelioides (Hook.) Tuck. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 80. 1867.

Not as usually cited: (Hook.) Trev., Lichenoth Ven. 98. 1869.

Basionym: *Lecidea parmelioides* Hook. in Kunth, Syn. Pl. Itin. Plag. Aeq. Orb. Nov. **1**: 15. 1822.

At least in its broad sense, this species ranges widely in the milder regions of the northern hemisphere. In the manuscript Tuckerman had added as additional new combinations two varieties to this species, but they were dropped from the publication by Curtis.

Enchylium polycoccum (Nyl.) Tuck. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 77. 1867.

Basionym: *Synalissa polycocca* Nyl., Syn. Meth. Lich. **1**: 96. 1858.

This species, now classified as *Pyrenopsis polycocca* (Nyl.) Tuck., is known only from the United States.

Heppia despreauxii (Mont.) Tuck. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 80. 1867.

Not as usually cited: (Mont.) Tuck., Gen. Lich. 46. 1872.

Basionym: *Solorina despreauxii* Mont. in Barker-Webb & Berth., Hist. Nat. Iles Canaries 3, 2ème Part., Sect. 3. 104. 1840.

This species, now known as *Heppia lutosa* (Ach.) Nyl., occurs at least in North America and Europe.

Lecanora myrini (Th. Fr.) Tuck. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 80. 1867.

Not as usually cited: (Fr.) Nyl., Flora **52**: 413. 1869.

Basionym: *Aspicilia cinerea* γ *myrini* Th. Fr., Lich. Arctoi [=Nova Act. Reg. Soc. Sci. Upsal., ser. 3, 3]. 132 [not "231"]. 1860.

The basionym of this species has previously been thought to be *Parmelia myrini* Fr. (Sum. Veg. Scand. 1: 108. 1848) which is actually a *nomen nudum*. Apparently Th. Fries was the first to describe the species (see above) and therefore he and not E. Fries must be considered the author. And Nylander's incorporation of this species into the genus *Lecanora* is antedated by Tuckerman's new combination.

This species is known from North America, northern Asia, and Europe.

Urceolaria lepadina (Ach.) Tuck. ex M. A. Curt., Geol & Nat. Hist. Surv. N. C. **3**: 80. 1867.

Basionym: *Lichen lepadinus* Ach., Lichenogr. Suec. Prod. 30. 1798.

Although the name of this species appears in both the manuscript and the publication as "*Urceolaria lepadina* (Sommerf.)," there is no doubt that the Acharian basionym is meant as it is the only "lepadinum" in a genus even remotely related to *Urceolaria*. The range of *Thelotrema lepadinum* (Ach.) Ach., as this widely distributed species is now classified, surely includes North Carolina, but a certain doubt is cast on the identification inasmuch as the species is recorded from rock rather than bark. Tuckerman, however, probably did not see a specimen because he added in the manuscript "Schweinitz in herb. Fries," apparently inserting the name on Schweinitz's authority. For nomenclatorial purposes, however, the accuracy of the determination is not significant.

To his original description of *Lichen lepadinus Acharius* (see above) added as a synonym "*Urceolaria ? lepadina*." This name, published as a synonym, has no nomenclatorial status.

2. Nomina Nuda for Species Later Described

Arthonia glaucescens Nyl. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 82. 1867 = *A. glaucescens* Nyl. in Willey, Syn. Gen. Arth. 20. 1890.

This species is known only from North America. In Tuckerman's manuscript, but not in the publication, it is remarked that this species was "determined, as all the other species of the genus, and many others of the tribe [Graphidei], by Dr. Nylander."

Graphis erumpens Nyl. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 81. 1867 = *G. erumpens* Nyl. in Hue, Nouv. Arch. Mus. [Paris], sér. 3, **3**: 158. 1891.

This species, now classified as *Phaeographis erumpens* (Nyl.) Müll. Arg., is known only from North America.

Lecidea recedens Nyl. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 81. 1867 = *L. recedens* Nyl. in Hue, Nouv. Arch. Mus. [Paris], sér. 3, **3**: 135. 1891.

This species, known only from North America, is excluded from the Lichenes as a fungus by Zahlbruckner (Catal. Lich. Univ. **3**: 896. 1925).

Lecidea schweinitzii Fr. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 81. 1867 = *Biatora schweinitzii* Tuck., syn. N. A. Lich. **2**: 43. 1888.

Bacidia schweinitzii (Tuck.) Schneid., as this species is now known, ranges widely in eastern North America. Elias Fries, Tuckerman's most trusted mentor in lichenology,

supplied the name. In the publication the name appears only as *Lecidea schweinitzii* Fr., but in the manuscript it is placed in the section Biatora, thereby leaving no doubt of the intent. The attributing of the later described *Biatora schweinitzii* to Tuckerman must have been done by Willey, who edited the posthumous publication in which it appeared. In the absence of additional information, there is no justification for changing this authority.

3. Nomina Nuda

The following six species were apparently never described, at least not under the names given here.

Coniocybe albella Schwein. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 77. 1867.

The manuscript adds "Schwein. in hb. Fries," surely indicating that Tuckerman saw no specimen but put in the name on Schweinitz's authority.

Lecidea absconsa Tuck. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 81. 1867.

This is the only one of the unlisted names that does not appear in Tuckerman's manuscript.

Myriangium curtisii Mont. & Berk. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 77. 1867.

Pyrenastrum simplex Ravenel ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 83. 1867.

After Ravenel's name in the manuscript is the abbreviation "ms." and the insertion of *Verrucaria duplicans* Nyl. as a synonym. It seems doubtful, however, that the tropical *Anthracothecium duplicans* (Nyl.) Müll. Arg. occurs in North Carolina.

Verrucaria spinulosa Schwein. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 82. 1867.

The manuscript adds "hb." after this name.

Verrucaria thelomorpha Tuck. ex M. A. Curt., Geol. & Nat. Hist. Surv. N. C. **3**: 82. 1867.

The manuscript adds "ms." after this name.

LITERATURE CITED

CULBERSON, W. L. 1960. The Collected Lichenological Papers of Edward Tuckerman. H. R. Engelmann (J. Cramer), Publisher. Weinheim/Bergstrasse.

CURTIS, M. A. 1867. Geological and Natural History Survey of North Carolina. Part III, Botany; Containing a Catalogue of the Indigenous and Naturalized Plants of the State. Raleigh, N. C. 158 pp. [Lichenological part covers pp. 75–83.]

TUCKERMAN, E. 1866. A letter from Amherst, Mass., to M. A. Curtis, March 25, 1866. Southern Historical Collection, University of North Carolina Library, Chapel Hill.

WILLEY, H. 1886. Bibliographical sketch [of Tuckerman's works]. Bot. Gaz. **1886**: 74–78.

THOMAS JEFFERSON'S LIBRARY OF APPLIED CHEMISTRY

HAROLD J. ABRAHAMS
Germantown, Pa.

Thomas Jefferson was a scientist turned president. There is scarcely a science of his day which did not command his interest, and in certain sciences, such as paleontology, he was a pioneer. Time for research being unavailable to a person in his position, he nevertheless lent his enormous influence and encouragement to those contemporaries who did devote their time and talents to research in the various sciences.

Of great interest to him were the *applications* of science, especially those related to the affairs of daily life, and thus useful to the many, rather than the few. Many letters exist which bear Jefferson's congratulations to a scientist or layman for his practical contribution to domestic chemistry or for his work in disseminating knowledge of these practical applications in a manner comprehensible to the public.

To Thomas Cooper, professor of chemistry at Carlisle College, July 10, 1812:

> . . . . I have just entered on the reading of it [the Introductory Lecture of Thomas Cooper, Esq.] and perceive that I have a feast before me. . . . You know the just esteem which attached itself to Dr. Franklin's science, because he always endeavored to direct it to something useful in private life. The chemists have not been attentive enough to this. I have wished to see their science applied to domestic objects, to malting for instance, brewing, making cyder, to fermentation and distillation generally, to the making of bread, butter, cheese, soap, to the incubation of eggs, etc. And I am happy to observe some of these titles in

the syllabus of your lecture. I hope you will make the chemistry of these subjects intelligible to our good housewives. . . .

To Thomas Ewell, August 30, 1805:

. . . . of the importance of turning a knolege of chemistry to household purposes I have been long satisfied. the common herd of philosophers seem to write only for one another. the chemists have filled volumes on the composition of a thousand substances of no sort of importance to the purposes of life; while the arts of making bread, butter, cheese, vinegar, soap, beer, cyder, etc. remain totally unexplained. Chaptal has lately given the chemistry of wine making. the late Doct. r Pennington did the same as to bread, & promised to pursue the line of rendering his knoledge useful to common life; but death deprived us of his labors . . . .

To Caspar Wistar, January 4, 1805: ". . . . Morveau's Sur les moyens de desinfecter l'air . . . is a work of great interest to cities subject to infection, to hospitals, vessels & indeed to the country inhabitants. if you have not seen it, I will send it to you. . . ."

Again to Caspar Wistar, November 2, 1806, he wrote that he was depositing Morveau's book and a set of his permanent and portable apparatus for disinfecting air with the American Philosophical Society: ". . . . they may be able, thro' their Medical members especially, to ascertain by experiments the efficiency of M. de Morveau's process. . . . I conceived that his process might become of great public value. I considered it with a view to the disinfection of vessels from suspected ports. . . ."

To Monsieur L'Hommande, August 9, 1787:

. . . . Every discovery which multiplies the subsistence of man, must be a matter of joy to every friend of humanity. As such, I learn with great satisfaction, that you have found the means of preserving flour more perfectly than has been done hitherto. But I am not authorized to avail my country of it, by making an offer for its communication . . . . We are only to hope, therefore, that those governments who are in the habit of directing all the actions of their subjects, by particular law, may be so far sensible of the duty they are under of cultivating useful discoveries, as to reward you amply for yours, which is among the most interesting to humanity. . . .

While he was American minister to France his inspections of many European industrial and agricultural establishments engaged in the preparation of wines, olive oil, and dairy products, or in mailing grain or curing hay, etc., indicate his ever present interest not only in applied science but also in the welfare of his countrymen, on whose behalf he made these studies.

He himself never took out any patents, but gave away freely his ideas on the plow, the dry dock, the hemp machine, and so on, believing that his reward lay in their benefit to mankind.[1] Yet when his duties as Secretary of State in President Washington's cabinet included administering the new patent law, he recognized the propriety of rewarding the ingenuity of others whose applications of science produced methods of making life easier and more comfortable. He opposed monopoly as inimical to the technological progress of the new nation, but he tried to protect an inventor's rights and even argued that a period of fourteen years was not adequate protection for an inventor in such a large country.

Before granting a patent he tried out the invention either by himself or in company with a group of experts; note the application by Jacob Isaacs for a patent on a process for making fresh water from salt water. In the first year of the patent law he granted only three patents, and until the change in the law in 1793 the total number granted was very small. However, despite Jefferson's opposition, in that year the law was liberalized, presumably because of his strict interpretation of it. Nevertheless, Jefferson was much pleased by the increase of inventions after the passage of the patent law, and he saw this technological advance as a bulwark of the sound, democratic—and hence moral—way of life.

The emphasis of his excellent book *Notes on the State of Virginia* on the various resources of his native state reflects his preoccupation with applications of chemistry and other sciences. He devotes thirteen pages to Virginia's deposits of gold, lead, copper, iron, graphite, coal, precious stones (emerald and amethyst), marble, limestone, stone (millstone, slate, flint, mica, magnetite, asbestos), earths (clays, chalk, gypsum, and ochre), nitre, salt and medicinal springs.

His *Farm Book* shows his application of chemistry to agriculture in general and his attention to such processes as fermenting, brewing and distilling, and tanning, and to gunpowder, building materials (such as stone, lime, glass, paint), coal and charcoal, and so on. Jefferson's plantation at Monticello was vast and rather isolated. He had therefore to strive

[1] Nevertheless he awarded Oliver Evans, inventor of machinery which Jefferson used in his mill, a sum of money as an honorarium to a person from whose ingenuity others benefited.

for a certain self-sufficiency for his family, domestic servants, slaves and tenants.

Some animal hides came from his own cattle and some he bought. Part of these were probably tanned at Monticello. A record in the *Farm Book* shows that Jefferson ordered his men to "Get bark for tanning leather where your next clearing is to be, felling the tree & stripping it clean." The leather was used for making shoes, harnesses and saddles, for there was great demand for such items. In 1794 alone, sixty pairs of shoes were required for his slaves.

Until 1795 he bought the charcoal used as fuel for his home and for the smith shop. In that year he began to make his own, for which he had four kilns.

The "Account Book, 1799," has these items:

May 14. I am always to give Frank a half dime for every bushel to the cord of wood which his coal kiln's yield. his last yielded 30 bushels to the cord: therefor paid 1.5 D.

Nov. 30. pd Frank for his 4th kiln 33. half dimes + a bottle of molasses @ ⅓ note it yielded 37. bush. to the cord.

At his plantation called Limestone on the Hardware River in Albemarle County, limestone was quarried, then transported to Monticello and converted into lime in kilns.

His "Account Book, 1771" reads:

July 31. Bishop has worked 3½ days for which am to give him 2/6. he will undertake to mould and burn bricks for 7/6 the 1000. and requires two men, three boys, and no more he counts those that are worked up. but better give him £3.10 a month.

Sep. 11 Stephen Willis sais it takes 15. bushels of lime to 1000 bricks.

Sep. 16 Defoe begins to blow lime @ 30/per month Will Beck to find him provns for which allow 15/per month

Jefferson's workmen used much gunpowder in his extensive building operations. Local merchants sold powder of such poor quality that he often had to send for a better grade to the distant firm of Dupont in Wilmington. At Monticello he had an eprouvette for testing the powder and he was thus able to estimate its power before it was used.

To Patrick Gibson, from Monticello May 20, 1793:

. . . . I have the eprouvette used in our war department for proving powder. dupont's carries that to 27. however 22. or 23. is receivable at the War office . . . . the first barrel you sent me was at 9. the last at 7. it is not merely the difference in quantity which might be thought requisite to make up the difference of force: but in boring rock, even if you were to treble the size of the hole & of course the expense of the boring, yet such a quantity of powder would not at all kindle but be blown out unburnt. but this weak powder cannot force out more of the rock than barely spoils the hole bored . . .

Jefferson himself was fond of wines and cider. Whiskey, rum, and ale were for the use of workmen and slaves when engaged in certain kinds of work only, and under other extenuating circumstances . . . "when they work in the water in cool weather . . . which might produce colds . . . colds & sickness." Thus in 1813 he began to brew liquors on his plantation. His distillery was set up by Captain Joseph Miller, a London brewer who was confined to Albemarle County for political reasons during the War of 1812. Miller and Jefferson became good friends and he lived at Monticello for a time, and from him the household servants learned how to malt and brew. Fall brewing was done in October and November and spring brewing in April and May, using corn, rye, and wheat.

In a letter to George Divers, November 26, 1792, Jefferson requested information on what kind of stills to buy. Divers advised one still of about 85 gallons but not over 100 gallons and another of about 45 but not over 70 gallons, and further, that "a pewter worm is better than a copper." Such equipment, he wrote, would enable Jefferson to make ". . . from 70 to 80 gallons of whiskey p. week & feed 60 or 70 Hogs, the feeding that number of Hogs will be an object to you as it will save a considerable quantity of Indian Corn, from which consideration I have been induced to recommend it to you to purchase stills of the above size, that you may be able to keep them at work for 7 months in the year in which time they will Distill about 900 Bushels of grain . . . ." (The *Farm Book* also contains a drawing of an apple mill and press by Jefferson.)

He wrote requesting the purchase of a copy of *The American Brewer and Maltser* by Joseph Coppinger on September 18, 1813, having seen it advertised in the *Aurora*. To Coppinger he wrote (April 25, 1815): ". . . . I am lately become a brewer for family use, having had the benefit of instruction to one of my people by an English brewer of the first order." After declaring that he had "perfect success" with Coppinger's book in malting corn the previous fall, he said that although the brewers on the James River used corn almost exclusively during the Revolutionary War, he used wheat.

To Captain Miller he wrote on June 26,

1815, that Peter Hemings had brewed some corn which would have been good had he not spoiled it by overhopping, but that he would become a good brewer with experience. There are letters which show Jefferson trying to purchase jugs, corks, etc. To James Madison he wrote that the person whom Madison was to send to Monticello during the brewing season in October would be given many successive lessons in brewing before his return to Madison.

> . . . try his hand . . . . discover what parts of the process he will have learnt imperfectly and come again to our spring brewing of a single cask in order to perfect himself, and go back to you and try his hand again on as much as you will want.

He then gave Madison advice on the equipment needed for this enterprise.

Jefferson once thought that when he retired he would rehabilitate his wasted lands, and that since his vast woodlands were not suited to farming and too far from his farms for the timber to be used there, the wood might be burned and crude potash manufactured. Some of the product would be used as fertilizer on his lands and the remainder sold. He wrote to Monsieur de Meusnier on April 29, 1795, that such an activity ". . . requires but small advances of money. I concluded at length however to begin a manufacture of nails which needs little or no capital."

To Joseph Leacock he wrote on November 24, 1792, reviewing certain facts and figures about conversion of wood potash and telling why the process interested him. Leacock's return account of the difficulties involved was discouraging.

Jefferson's library gives testimony to his interest in the application of chemistry to the service of man. He scoured Paris for books dealing with all of the sciences, and on his return to America estimated that he owned "what is chiefly valuable in science and literature"—between nine and ten thousand volumes. This Jefferson library, the inventory of which is known as the 1815 Catalogue, was purchased by the United States government and formed the nucleus of the Library of Congress.

The Library of Congress owns thirty Jefferson books on pure chemistry. In addition there are at least forty works on applied chemistry, scattered among other headings as follows: [The symbols 4 to, 8 vo, and 12 mo indicate quarto (about 8 by 11 inches), octavo (6 by 9), and duodecimo (4 by 7), respectively.]

Natural Philosophy

"In Natural Philosophy I mean to include Chemistry and Agriculture."—Jefferson's letter to J. Banister, Jr., from Paris dated October 15, 1785.

1. MITCHILL, S. L. *Outline of the Doctrines in Natural History, Chemistry, and Economics.* New York, 1792. 8 vo. Mitchill (1764–1831) was a New York physician of profound learning who corresponded with Jefferson for a number of years. He was professor of chemistry at Columbia College.

2. ROUELL, JOHN. *A Complete Treatise on the Mineral Waters of Virginia . . . Analysis, Contents, ...* Philadelphia, 1792. 8 vo.

3. RUSH, BENJAMIN. *An Account of the Sugar Maple Tree of the United States and the Methods of Obtaining Sugar from It . . . .* Philadelphia, 1792. 8 vo. A footnote on page 14: "Mr. Jefferson uses no other sugar in his family, than that which is obtained from the sugar maple tree. He has lately planted an orchard of sugar maple trees on his farm in Virginia."

Rush (1745–1813) was the famous Philadelphia physician and close friend of Jefferson.

4. CONOVER, S. F. "An Essay on the Vermilion Color of the Blood . . . ." Unpublished manuscript. 1809. Another Philadelphia physician.

A presentation copy, probably in manuscript, from the author. Jefferson replied on December 23, 1807: "Th: Jefferson presents his compliments to Doct.r. Conover and his thanks for the memoir he has been so kind as to inclose him, and which he has read with satisfaction. it is always pleasing to see the discoveries in science applied to the advancement of the arts, or of the useful or ornamental purposes of life."

Agriculture

5. BRADLEY, RICHARD. *Ten Practical Discourses Concerning Earth and Water, Fire and Air, as they Relate to the Growth of Plants.* London, 1733. 8 vo. Bradley (1688–1732) was an English horticultural author.

In June, 1781, Jefferson put together his great accumulation of data on his native state and published *Notes on the State of Virginia.* This was the first comprehensive book on any section of the United States and, measured by its influence, was the most important American scientific work to that date. In it can be seen the influence of Bradley's book. "It is by the assistance of heat and moisture that vegeta-

bles are elaborated from the elements of earth, air, water, and fire," Jefferson wrote in 1782, before Lavoisier's new ideas had gained currency.

6. SPURRIER, JOHN. *The Practical Farmer . . . Containing the . . . Chemical . . . Elements of Agriculture.* Wilmington, Del., 1793. 8 vo. (dedicated to Jefferson).

In connection with this book Jefferson wrote, on December 29, 1794:

> . . . the horse bean I tried this last year. it turned out nothing. the President has tried it without success. an old English farmer of the name of Spuryear settled in Delaware, has tried it there with good sucess: but he told me it would not do without being well shaded, and I think he planted it among his corn for that reason. but he acknoleged our pea was as good an ameliorater & a more valuable pulse, as being food for man as well as horse. . . .

7. HALES, STEPHEN. *Statical Essays . . . Also a Specimen of an Attempt to Analyse the Air . . . .* London, 1738. 8 vo. 2 vol.

Hales (1677–1761), "the father of vegetable physiology," was an English clergyman, scientist, and inventor.

8. KIRWAN, RICHARD. *The Manures most advantageously applicable . . . .* London, 1796. 8 vo.

Jefferson wrote to William Strickland on March 23, 1798:

> . . . I am much indebted to you for Mr. Kirwan's charming treatise on manures. science never appears so beautiful as when applied to the uses of human life, nor any use of it so engaging as those of agriculture & domestic economy. Doct.r Home had formerly applied the doctrines of chemistry to the analysis of soils & manures, but the revolution in that science had required the work to be done over again, and gives to Mr. Kirwan the entire merit of a new work.

Kirwan (1733–1812) was an Irish chemist and member of the Royal Irish Academy, Royal Society, and American Philosophical Society.

9. HOME, FRANCIS. *The Principles of Agriculture and vegetation.* London, 1762. 8 vo.

Home (1719–1813) was a professor of materia medica at Edinburgh University. *The Principles* was a prize essay and went through numerous reprintings and translations.

10. GYLLENBORG, GUSTAVUS ADOLPHUS. *The Natural and Chemical Elements of Agriculture.* Translation by John Mills. London, 1770. 12 mo.

Gyllenborg (1743–1789) was a Swedish banker.

11. PETERS, RICHARD. *Agricultural Enquiries on Plaister of Paris.* Philadelphia, 1797. 8 vo.

Jefferson wrote on March 6, 1816: ". . . we are indebted to you for much of our knowledge as to the use of the plaister, which is become a principal article of our improvements, no soil profiting more from it than that of the country around the place . . . ."

Peters (1744–1828) was a friend of Jefferson's and a lawyer, patriot, and farmer. *Agricultural Enquiries* was written at the request of George Washington.

12. CHAPTAL, JEAN A. C., FRANÇOIS ROZIER, ANTOINE A. PARMENTIER, AND LOUIS D'USSIEUX. *Traite theorique et practique sur la Culture de la Vigne, avec l'Art de Faire le Vin . . . .* Paris, 1801. 8 vo. 2 vol.

Jefferson wrote on December 16, 1809: ". . . the best ever published, on the vine, & on wines."

Chaptal, comte de Chanteloup (1756–1832) was a chemist, and Rozier (1734–1793) an agriculturalist. Parmentier (1737–1813) was a chemist and agriculturalist whose discoveries about the potato were so helpful to France that a proposal was once made that the French word for potato be "parmentière." D'Ussieux (1744–1805) was a writer.

Medicine

"Law, medicine, chemistry, mathematics, every science has a language of its own, and divinity not less than others."—Thomas Jefferson.

13. BAUME, ANTOINE. *Elements de Pharmacie . . . par les principes de la Chymie . . . .* Paris, 1784. 8 vo.

Baume (1728–1804) was a well-known French pharmacist.

14. MACBRIDE, DAVID. *Experimental Essays: I—On the Fermentation of Alimentary Mixtures; II—On the Nature and Properties of Fixed Air; . . . . V—On the Dissolvent Power of Quick-Lime.* London, 1764. 8 vo.

MacBride (1726–1778) was an Irish chemist and physician.

15. MCCLURG, JAMES. *Experiments upon the Human Bile . . . .* London, 1772. 8 vo.

McClurg (1746–1823) held the first chair of Anatomy and Medicine at the College of William and Mary.

16. WOODHOUSE, JAMES. *An Inaugural Dissertation on the Chemical and Medical Properties of the Persimmon Tree . . . .* Philadelphia, 1792. 8 vo. (a presentation copy by the author).

Jefferson wrote his thanks on May 31, 1792, for the ". . . interesting dissertation on the persimmon tree. he is happy to find that this plant may become a valuable addition to our stock of Chemical & Pharmaceutical subjects,

in both which lines Dr. Woodhouse has presented very interesting experiments on it. Th: Jefferson has for some time turned his attentions to the same plant as a subject of Agriculture . . . and he has been well informed that the quantity of Spirit it yeilds is considerably more than that mentioned . . . that it might be a substitute for the bark in tanning, is new to Th: J and adds new value to it . . . ."

Woodhouse (1770–1809) was a Philadelphia chemist and physician who taught chemistry at the University of Pennsylvania.

17. The Edinburgh New Dispensatory: containing *I—The Elements of Pharmaceutical Chemistry, . . . enriched by the latest discoveries in . . . chemistry* . . . . Philadelphia, 1791.

Mineralogy

18. SHORT, THOMAS. *The Natural, Experimental and Medicinal History of the Mineral Waters of Derbyshire . . . with . . . Experiments.* . . . London, 1734. 4 to.

Short (1690–1772) was an English physician.

Technical Arts

19. RUMFORD, BENJAMIN THOMPSON, COUNT. *Essays, Political, Economical and Philosophical.* Boston, 1798. 8 vo. 2 vol. (contains chapters on American food and cookery).

Rumford (1753–1814) was an American physicist and inventor. He founded the Royal Institution of Great Britain. Besides American food and cookery, his writings dealt with fireplace construction and other subjects. His observations on the heat generated during the boring of cannon later lead to an understanding of the mechanical equivalent of heat by Joule. He was also a military expert.

20. BERTHOLLET, CLAUDE LOUIS. *Elements de l'Art de la Teinture.* Paris, 1791. 8 vo. 2 vol.

Jefferson mentions Berthollet's work on fulminating powder and on bleaching in letters to Bishop Madison and to Doctor Currie. Berthollet (1748–1822) was a French chemist.

21. *Smith's Distillery.* Philadelphia. 8 vo.

22. KRAFFT, MICHAEL. *The American Distiller* . . . . Philadelphia, 1804. 8 vo.

A gift from the author and dedicated to Jefferson. Jefferson wrote on April 29th, 1804:

> I see too with great satisfaction every example of bending science to the useful purposes of life. hitherto chemistry has scarcely deigned to look to the occupations of domestic life. when she shall have made intelligible to the ordinary householder the philosophy of making bread, butter, cheese, soap, beer, cyder, wine, vinegar, etc. these daily comforts will keep us ever mindful of our obligation to her. the art of distilling which you propose to explain, besides its household uses, is valuable to the agriculturalist, as it enables him to put his superflous grain into a form which will bear long transportation to markets to which the raw material could never get.

Krafft, a native of Pennsylvania, was born about 1775.

23. MAUPIN. *Art de faire le Vin.* Paris, 1779. 12 mo.

24. CHARPENTIER DE COSSIGNY, JOSEPH. *Observations sur: L'art de faire le vin par M. J. A. Chaptal.* Paris, 1807. 8 vo.

Charpentier de Cossigny (1730–1809) was a French naturalist.

25. CHARPENTIER DE COSSIGNY, JOSEPH. *"Recherches physiques et chimiques sur la Fabrication de la Poudre à Canon etc."* Paris, 1808. 8 vo. 2 vol. (a presentation by the author).

26. LAVOISIER, ANTOINE LAURENT. *The Art of Manufacturing Alkaline Salts and Potashes.* Translated by Charles Williams. 8 vo.

This is of course Lavoisier (1743–1794), the great French chemist and "father of modern chemistry."

27. HOPKINS, SAMUEL. *Address to the Manufacturers of Pot and Pearl Ash.* New York, 1791. 8 vo. (a presentation copy by the author).

28. *Remarks on the Manufacturing of Maple Sugar* etc . . . . "Collected by a Society Gentleman." Philadelphia, 1790. 12 mo.

29. RICHARDSON, JOHN. *Philosophical Principles of the Science of Brewing.* London, 1788. 8 vo.

Richardson lived in England, in the late eighteenth century. He was the first to write scientifically on brewing and introduced the saccharometer to brewers.

30. KNIGHT, THOMAS A. *A Treatise on the Culture of the Apple & the Pear and on the manufacture of Cider and Perry.* London, 1802. 12 mo.

Knight (1759–1838) was an English vegetable physiologist, president of the Horticultural Society, and fellow of the Royal Society.

31. EALES, MARY. *The Compleat Confectioner; or the Art of Candying and Preserving in its utmost perfection.* London, 1742. 12 mo.

Mistress Eales lived in the early eighteenth century and was confectioner to Queen Anne. The first edition of this work is dated 1718.

32. PARMENTIER, ANTOINE A. (The following three titles have been translated and condensed.) *Advice . . . on . . . Best Way to Make Bread.* Paris, 1782. 12 mo.

33. PARMENTIER, ANTOINE A. *The Perfect Baker.* Paris, 1778. 8 vo.

34. PARMENTIER, ANTOINE A. *Manufacture of Syrups* . . . . Paris, 1812. 8 vo.

35. FABBRONI, ADAMO. *Dell 'Arte di fare il vino.* Firenze, 1787. 12 mo.

Jefferson wrote, on May 24, 1789, in thanks for this gift ". . . the subject of wine, a subject interesting to me, and which had not before been philosophically treated . . ."

Fabbroni was a Florentine agriculturalist.

36. *Manner of Making Tar in Sweden.* 4 to. (Author, place, and date not given.)

37. APICIUS, COELIUS. *Apicii Coelii de Opsoniis et Condimentis* . . . . Amsterdam, 1709. 8 vo.

This book on cookery was one of a select group which Jefferson ordered in Amsterdam. The receipts in it date from the third century, A. D. This edition was annotated by Martin Lister (1638–1712), the English zoologist.

38. FENNEL, JAMES. *Description of the Principles and Plan of Proposed Establishments of Salt Works . . . Supplying the U.S. with home made salt.*

A presentation copy from the author. Jefferson replied on June 12, 1798:

> . . . the theory is certainly promising . . . having no experience on the subject . . . he [Jefferson] . . . is entirely unable to give an opinion; but doubts not that Mr. Fennel has sufficiently verified his process by experiment . . . he sincerely wishes him success, and apprehends the circumstances of the times are likely to befriend his undertaking.

Fennel (1766–1816) was an English actor and playwright who was invited to come to America in 1793 by the manager of the Philadelphia Theater. The salt works which he set up near New London, Conn., was not financially successful.

39. ELLIS, WILLIAM. *The London and Country Brewer* . . . . London, 1750. 8 vo.

On September 17, 1813 Jefferson wrote to Captain Meriwether:

> . . . we are this day beginning, under the directions of Capt. Millar, the business of brewing Malt liquors, and if these books [i.e., Ellis and Combrune, No. 40 below] are no longer useful to you I will thank your for [returning] them as we may perhaps be able to derive some information from them.

Ellis lived in the first half of the eighteenth century and was an English agriculturalist.

40. COMBRUNE, MICHAEL. *The Theory and Practise of Brewing.* London, 1804. 8 vo.

On September 18, 1813, Jefferson wrote: ". . . I see . . . advertised . . . the American Brewer and maltser which, as teaching the method of malting Indian Corn I should be very glad to get. could you procure it . . . ."

On May 11, 1821 he wrote: ". . . I much doubt if the operations of malting & brewing could be successfully performed from a receipt. if it could Combrune's book on the subject would teach the best processes; and perhaps might guide to ultimate success with the sacrifice of 2 or 3 trials."

A mid-eighteenth-century man, Combrune's book was the first to recommend the use of a thermometer in brewing.

These constitute the applied chemistry books of the 1815 library which Jefferson sold to Congress in that year. His interest in the applications of chemistry will be obvious from the list.

Jefferson also had an earlier library (almost entirely destroyed by fire in 1770) and a later one, sold at auction in 1829. The Catalogue of the 1829 sale (by Nathaniel Poor) lists the following works on pure and applied chemistry:

1. Cooper, *Introduction to Chemistry.*
2. Lavoisier, *Traite.*
3. Marcet, *Conversations on Chemistry* (Cooper revision).
4. Brande, *Manual of Chemistry* (notes by Macnevin).
5. Bache, *Medical Chemistry.*
6. Kirwan, *Manures.*
7. Chaptal, Rozier, Parmentier, Ussieux, *Sur la Vigne et le Vin.*
8. Cadet de Vaux, *Sur le Vin.*

Thus much evidence has been presented that Jefferson regarded the sciences highly for the practical contributions they could make to the comfort and happiness of mankind. He hoped that the sciences would become the cornerstone of the American republic, and that its schools could do justice "to our country, its productions and its genius." Benjamin Franklin and John Adams displayed the same attitude toward the sciences, reasoning that a new country with immense natural resources needed a great corps of technologists, oriented toward swift development of our potentialities.

Jefferson's particular interest was applied chemistry. His detailed, signed order to Captain Meriwether Lewis (June 20, 1803) directed Lewis to give attention to "mineral productions of every kind, but particularly metals, limestone, pit-coal and saltpetre, salines and mineral waters . . .," and his counsel to Dr. Emmett at the University of Virginia placed great emphasis on applied chemistry. We would be unfair to Jefferson, however, if we failed to note that he could be a speculative scientist as well—witness his interest in the "megalonyx,"

and his thoughts on the existence of seashells on mountaintops. But his letters and books show that his interests centered around what was utilitarian to man, and it was this aspect of chemistry that merited his attention.

ACKNOWLEDGMENT

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BIBLIOGRAPHY

BETTS, EDWIN M. 1953. *Thomas Jefferson's Farm Book*. Princeton University Press: Princeton, N. J.

BROWNE, CHARLES A. 1944. *Thomas Jefferson, Chronica Botanica*, Vol. 8, 1944.

ROSENBACH, A. S. W. 1936. *A Book Hunter's Holiday*, Boston.

SOWERBY, E. MILLICENT. 1952. *Catalogue of the Library of Thomas Jefferson*, Vol. I, Library of Congress.

ADDITIONS TO THE FRESH-WATER ALGAE IN NORTH CAROLINA V[1]

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Our knowledge concerning the fresh-water algal flora of North Carolina has almost doubled in the past decade. Prior to 1950, about 725 species were known from the state. With the addition of the taxa reported in this paper the number is now over 1,300. Studies by Phillips and Whitford (1958), Schumacher and Whitford (1959), R. C. Smith (1958), and Whitford (1956 and 1958) have been mainly responsible for this increase.

This article represents material collected, for the most part in the last two years, from all areas of the state. It contains reports on ninety-eight taxa. Twenty-seven of these are extensions of previous records or notes of important observations. Seventy-one are new records for the state. This includes one variety new to science (*Staurastrum*), first reports for eighteen genera, and fifty-two species and varieties. The new genera are marked with an asterisk. The regions from which the taxa are reported are designated by the following abbreviations: M (mountain), P (Piedmont) and CP (Coastal Plain). When the abbreviations appear without parentheses, they designate the range as given by the current paper. When they appear within the parentheses, they give the range as reported by earlier studies.

The authors wish to thank Drs. Francis Drouet, M. A. Faridi, L. H. Flint, N. Islam, C. W. Reimer, and Richard Wood for their determination of certain algae; Mr. Arthur M. Scott for his excellent drawings and aid in the description of the new variety of *Staurastrum;* and Mrs. Katherine Edsall for the Latin diagnosis.

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Chlorophyceae

VOLVOCALES

Pteromonas sinuosa Chodat. In plankton of Neuse and Roanoke Rivers, June, Halifax and Wayne Counties, CP.

Eudorina unicocca G. M. Smith. In tow of Neuse River, Craven County, CP, (CP, P).

TETRASPORALES

Schizochlamys gelatinosa A. Braun. Appeared in dish in 4°C. cold room in water from Limestone Creek, Duplin County, CP, (M).

**Stylosphaeridium stipitatum* Geitler and Gimesi. Found on colonies of *Coelosphaerium naegelianum* in plankton of Kerr Reservoir; collected by G. Elkan, July 6, 1960, Vance County, P.

ULOTRICHALES

Cylindrocapsa geminella Wolle. On stems in quiet water of Turkey Creek, Moore County, CP, (P).

Stigeoclonium amoenum Kützing. With *Tuomeya fluviatilis* on rocks in rapids of Nector Creek, Harnett County, CP.

Stigeoclonium flagelliferum Kützing. On rocks in rapid, sandy brook, Johnston County and in a creek west of Elizabeth City, Pasquotank County, CP.

Stigeoclonium lubricum (Dillw.) Kützing. On stones along edges of Tar River, Nash County, CP.

Stigeoclonium lubricum var. *varians* (Hazen) Collins. Forming green tufts in rapids of Mountain Creek, Richmond County, CP, (CP).

Stigeoclonium pachydermum Prescott. Attached to rocks in rapids of Rocky Creek, Wake County, P.

Stigeoclonium tenue (Ag.) Kützing. On rocks in rapid water of Sleepy Creek, Wayne County, and Neuse River, Wake County. On *sparganium* leaves in swift current of White Oak River, Onslow County, CP, (P).

Stigeoclonium tenue var. *irregulare* Wolle. In Sleepy Creek, Wayne County, CP.

**Protoderma viride* Kützing. This species is widespread and frequently abundant in spring and summer. It has been collected only on rocks in the rapids of streams, CP, P, M. Studies of colonies on glass slides indicate the species is not a juvenile stage of *Stigeoclonium* as often suggested. *Protoderma* grows as distinct branching filaments without upright cells. Local species of *Stigeoclonium* start growth as a tight layer of angular cells which produce upright filaments very early.

Coleochaete pulvinata A. Braun. On rocks in Swift Creek, Wake County, P.

Chaetosphaeridium globosum (Nordst.) Klebahn. On mosses and glass bottle in quiet water of creek and pond, Wake County. In Ravenel Pond, Macon County, P, M.

* *Dicranochaete reniformis* Hieronymus. In littoral zone of Catherine Lake, Onslow County, July, 1959, CP.

Gongosira lacustris Brand. Forming parenchymatous spots on old shells along edge of Tar Rivèr at Grimesland, Pitt County, CP.

OEDOGONIALES

Oedogonium howardii G. S. West. In laboratory aquarium for the past five years; fruits frequently; Wake County, P.

Oedocladium lewisii L. Whitford. Fruiting specimens of this alga have been collected in Carteret, Craven, Lenoir, Orange and Wake counties in autumn and in Wake and Harnett counties in spring. Vegetative material has been widely collected in both spring and autumn in the CP and P; in the mountains it has been found in Ashe County in August. The species grows on moist soil in bottom-land fields and along streams, with moss protonema and thalloid liverworts. M, (CP, P).

CLADOPHORALES

Pithophora oedogonia (Mont.) Wittrock. In rapids of Neuse River, Johnston County, in farm pond in Granville County, CP. The Neuse River collection was previously identified erroneously as *Cladophora crispata* (Whitford, 1956).

CHLOROCOCCALES

**Errerella bornhemiensis* Conrad. In tow, Haw River, Aug., 1959, Chatham County, P. (Pascher, 1915).

Characium ambiguum G. M. Smith. Common on Hydrodictyon in pond, McDowell County, M. (CP).

Hydrodictyon reticulatum (L.) Lagerheim. This alga had been reported by Whitford (1956) from a single station in Wake County. In July, 1960, a second station was established when it was collected by Ray Smith in a pond near Marion, McDowell County, M (P).

**Trochisia granulata* (Reinsch) Hansgirg. In plankton of Neuse River, June, 1960, Wayne County, CP.

Excentosphaeria viridis G. T. Moore. In shallow rock pools on Whiteside Mountain, July, 1958, Macon County, M, (CP).

Oocystis elliptica W. West. Forming bloom in shallow water, Collie Swamp, July, 1958, Martin County, CP. (M).

Tetraedron constrictum G. M. Smith. In plankton of Kerr Reservoir, July, Vance County, P.

Scenedesmus anomalus (G. M. Smith) Ahlstrom and Tiffany. In nannoplankton, Haw River, Chatham County, P.

Scenedesmus producto-capitatus Schumla. Same location as above species, P.

ZYGNEMATALES

Mougeotia recurvata (Hassall) de Toni. Roadside ditch west of Longwood, Brunswick County, CP.

Mesotaenium macrococcum var. *micrococcum* (Kütz.) W. and G. S. West. On *Scytonema* filaments from seep area in high sandhills, Spout Springs, March, 1960, Harnett County, CP.

Gonatozygon kinahani (Arch.) Rabenhorst. Common in Myatt's and Olive Pond, Wake County, P. As suggested by Irenée-Marie (1938)) the species is easy to confuse with filamentous Zygnemataceae.

Euastrum gemmatum Brebisson. In tow. Yadkin River, Yadkin County, P.

Euastrum oblongum (Grev.) Ralfs. Rare in tow of Watauga River, Watauga County, M, (P).

Cosmarium turpinii var. *eximum* W. West. In brown ooze in shallows on sand, Neuse River, Johnston County, CP.

Micrasterias arcuata var. *expansa* (Bail.)

Nordstedt. In squeezings from Horse Pond, *p*H 5.0, temp. 36°C., Carteret County, CP.

Micrasterias depauperata var. *kitchelii* (Wolle) W. West. Same location as above species, CP.

Xanthidium armatum var. *mediolaeve* G. M. Smith. In bottom ooze at 2½ feet. Horse Pond, Carteret County, CP.

Xanthidium subhastiferum var. *towerii* (Cushman) G. M. Smith. Rare in net collection from Yadkin River, Yadkin County, P.

Staurastrum leptocladum Nordst. In nannoplankton of Trent River, Craven County, CP.

Staurastrum manfeldtii var. *fluminense* var. nov. Schumacher.

Dentibus basibus in 3 collectionibus disponsis, una sub quoque processu, quaque collectione 3 et 4 aut 2 et 3 dentes contente; apicis verrucis eadem mensura in arcu, primis ultimisque verrucis cuiusque arcus extra margonem projectis; terminale parte cuiusque processus leviter turgida et 4 longas graciles spinas ferente, eis spinis oppositis in angulo recto inter se. Cellulis sine processibus 41–43 μ longis, cellulis 90–96 μ latis, isthmo 10–11 μ.

Basal teeth arranged in three groups, one beneath each process, each group containing 3 + 4 or 2 + 3 teeth; apical verrucae of same size in an arc, the first and last of each arc projecting beyond the margin; terminal portion of each process, slightly swollen and bearing 4 long slender spines, the opposite at right angles to each other. Cells without processes 41–43 μ long; cells 90–96 μ wide; isthmus 10–11 μ. See Figures 1–3.

In plankton of Neuse, Roanoke and Tar Rivers. Becomes plentiful, especially in the Roanoke, during the month of June and in several collections it was the most abundant phytoplankter. CP. (Delponte 1876).

Arthrodesmus octocornis Ehrenberg. In plant squeezing from a blackwater pond, Carteret County. Previously reported (Schumacher, 1958) in *Utricularia* bladders, Craven County CP, (CP).

Charophyceae

CHARALES

Nitella opaca Agardh. Growing near the surface of the water in winter and found only in deeper water in summer, Ravenel Pond, 3,800 ft. elev., *p*H 6.6, Macon County, M, (P).

Euglenophyceae

EUGLENALES

Euglena spirogyra var. *fusiformis* Deflandre. In Collie Swamp, Martin County, CP.

Phacus helikoides Pochmann. In Price Canal, Quinerly, July, 1958, Pitt County, CP.

Phacus orbicularis Huebner. With the preceding species, CP.

COLACIALES

Colacium arbuscula Stein. On crustacea from pool in Crabtree Park, March, 1960, Wake County, P.

Xanthophyceae

HETEROCAPSALES

Chlorosaccus fluidus A. Luther. The species has been collected in typical form in Wake, Lenoir, Onslow, and Bertie County (Whitford, 1960). The last collection in the Cashie River at Windsor, March, 1960, extends the range nearly 100 miles north. CP.

HETEROCOCCALES

°*Pseudotetraedron neglectum* Pascher. This species were reported from Iowa (Prescott, 1931) but the presence of pyrenoids makes the identification doubtful. Our cells are typical in structure and lack pyrenoids and starch. Rare in plankton of Catawba River, March, Catawba County, P. Figure 13.

°*Dioxys* sp. In plankton, common on *Difflugia* cases, August, 1960, Hester's Pond, Zebulon, Wake County, CP. This plant does not correspond to any of the described species in Pascher's monograph (Pascher, 1939). Figure 5.

°*Harpochytrium apiculatum* Pascher. A plant which seems identical with this incompletely described species (Pascher, 1939) was common on *Difflugia* cases in the plankton of Hester's Pond, Zebulon, Wake County (CP), August, 1960. Figure 9.

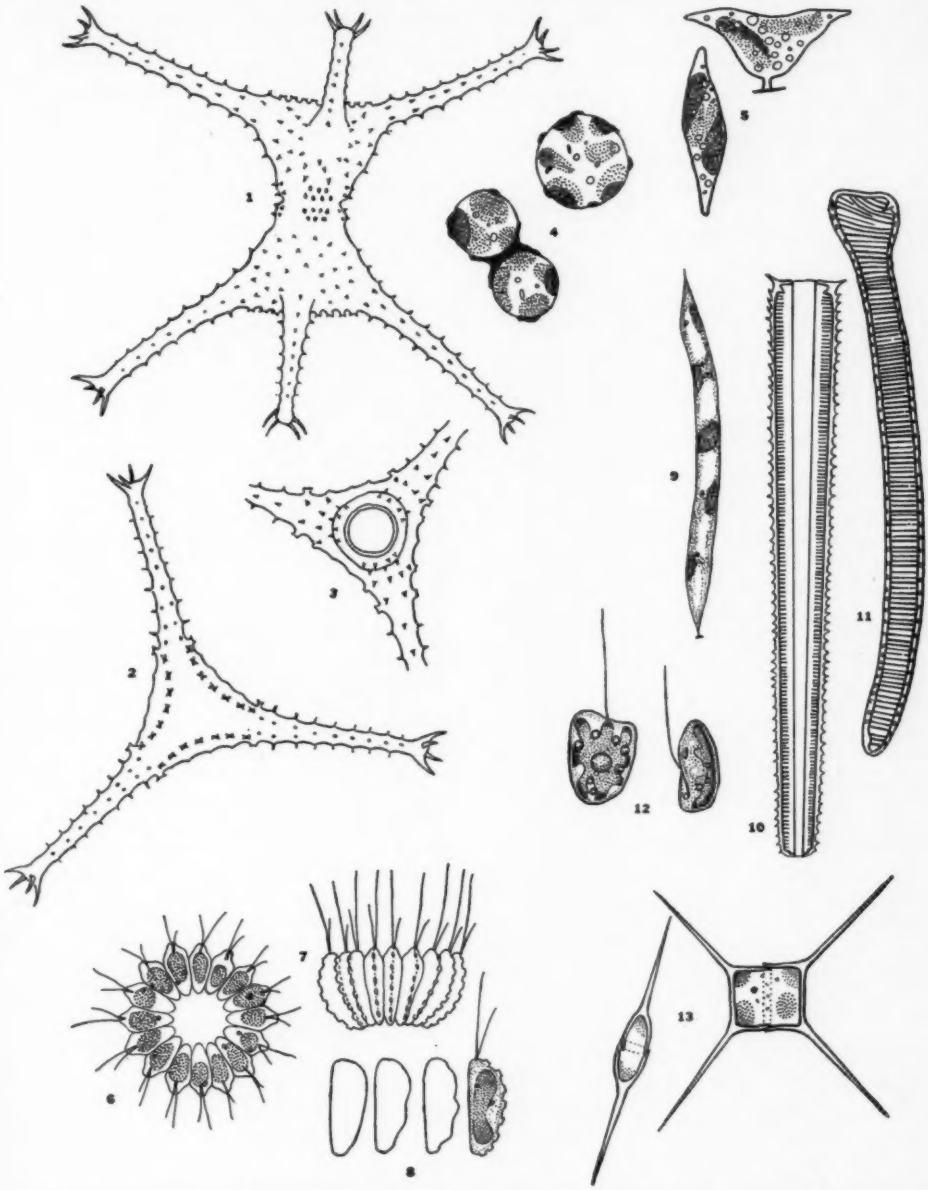
° *Chloridella cystiformis* Pascher. In temporary pool behind barracks, Mt. Mitchell, Yancey County, M. Elevation 2,000 meters, water temperature 10°C., March.

°*Chloridella neglecta* Pascher (1939). Abundant among filamentous green algae in a roadside brook near Garysburg, Northampton Co., March, 1960. The typical spherical cells (7–13 μ in diam.) frequently have spots or sheaths of brownish iron oxide. CP. Fig. 4.

Chrysophyceae

CHRYSOMONADALES

Chromulina nebulosa Cienkowski. This genus has been reported earlier by Whitford (1943) from the Coastal Plain, but a species determination was not made. Hoffman Wildlife Area Pond, Richmond County, CP.



FIGS. 1–13. 1. *Staurastrum manfeldtii* var. *fluminense* side view; 2. End view. 3. End view of isthmus. 4. *Chloridella neglecta.* 5. *Dioxys sp.* 6. *Cyclonexis annularis* end view of colony. 7. Side view of colony with papillate cells. 8. Side view of cells showing variation in shape. 9. *Harpochytrium apiculatum.* 10. *Actinella punctata* girdle view. 11. Face view (cells shorter than typical). 12. *Monochrysis aphanaster* face and edge views. 13. *Pseudotetraedron neglectum* edge and face views. (Figure 12 after Skuja, Figure 13 after Pascher.)

**Monochrysis aphanaster* Skuja (1948). In seep along Sleepy Creek, January, 1959, Wayne County, CP. Figure 12.

Microglena punctifera Ehrenberg. In seep along Sleepy Creek, January, 1959, Wayne County, CP, (P).

Mallomonas alpina Pascher and Ruttner. In farm pond, Newton Grove, Sampson County, CP.

* *Ochromonas mutabilis* Klebs. Abundant in seep near road, Sleepy Creek, March, Wayne County, CP.

Cyclonexis annularis Stokes 1886. (*C. uraliensis* Pochman 1957) The problem of this rare species, which is occasionally collected, is a puzzling one. It has been recently discussed at some length by Lund (1960) and Bourrelly (1957). Jane (1940) described a second species which differs in cell shape and colony form, and in 1957 Pochman described a plant (*C. uraliensis*) the cells of which have undulations on the peripheral wall.

On January 25, 1959 we collected *Cyclonexis* in a small seepage pool in Wayne County, North Carolina. The cells were somewhat like Pochman's plant—that is, they had undulations, which were almost papillae, on the peripheral wall. The cell has a single curved chromatophore which is sometimes rolled at the edges, as figured by Jane. The next winter (February, 1960) *Cyclonexis* was again collected in a small pool (Richmond County) and in a swamp pool (Moore County). In these collections the cell shape varied from the smooth cells figured by Stokes and Whelden (1939) to undulate cells similar to those of Pochman's plant. In the same collection were colonies having cells with a distinct row of papillae along the peripheral edge. There was only a single curved chromatophore with the curve inward, as figured by Jane. In peripheral view the cells were distinctly flattened and wedge shaped, with a distinct keel which might be smooth, undulate, or papillate, in the various colonies. The cells were blunt at the apex where the two unequal flagella are inserted.

It is our opinion that *C. uraliensis* is a variant of Stokes' species and should be placed in synonomy with it. *Cyclonexis erinus* Jane, on the other hand, with its truncate cells, distinctly flattened parallel to the colony rim, seems clearly a distinct species. If one does not accept this disposition of the variants, our plant with the papillate cells should be regarded as a species.

Algae such as *Cyclonexis* which apparently occupy very restricted micro-habitats, would be much more frequently collected if phycologists knew exactly when and where to look for them (Whitford, 1960). Our collections have all been made from acid pools (*p*H 5.0–5.4) in sandy soil in January and February, (water temperature 7–10°C.). The colonies occur sparingly in organic debris and ooze on the bottom. Figures 6 to 8.

Dinobryon utriculus Stein. In brown pool in swamp, Scotland County, CP, (P).

Hyalobryon deformans Lemmermann. This genus has been reported earlier by Whitford (1943), but no determinations to species were made. In brown pool in swamp, Scotland County, CP.

Hyalobryon ramosum Lauterb. Abundant colonies on water plants in 1 or 2 feet of water, Ravenel Pond, Macon County, March. In pool beside Shoeheel Creek, Scotland County, M, CP, March. (Cases of this species stain very nicely with Loeffler's Methyline blue in preserved material).

RHIZOCHRYSIDALES

Lagynion reductum Prescott. On *Melosira* filaments in Neuse River, Johnston County, CP.

CHRYSOCAPSALES

Phaeosphaera perforata L. Whitford (1943). Four recent collections have greatly extended the known range of this winter species. A collection in a brook near Highlands Biological Station in the mountains, Mar. 28, 1959, extends the range nearly 300 miles west. A collection in the Cashie River, March 21, 1960, in Bertie County, CP, extends it about 100 miles north in the state. Two out-of-state collections have also been made. One was in Florence County, South Carolina, February 21, 1960, and a second is near Lakewood, Ocean County, New Jersey, April 15, 1960 (F. H. Lesser, coll.). The known range now extends from Hattiesburg in southern Mississippi to New Jersey. It is found only in slow-flowing, generally brown-water streams at temperatures below 10°C., January to April.

Bacillariophyceae

CENTRALES

Melosira distans forma *clinostriata* Cleve-Euler. Common in summer nannoplankton, Neuse River, Johnston County, CP.

Melosira granulata var. *angustissima* forma *spiroides* Muller. Abundant in tows of the alkaline Roanoke River, especially during summer months, Halifax and Martin counties, CP. C. W. Reimer writes that this form has been collected only a few times in this country, and always in streams.

Melosira nummuloides (Dillw.) C. A. Agardh. In New River, Snead's Ferry, Onslow County, CP.

Cyclotella stelligera A. Cleve. and Grunow. In plankton of Neuse River, Wake County, P, (P).

Biddulphia laevis Ehrenberg. Living cells were collected in the plankton from several stations along the Tar River. (CP, P).

Hydrosera triquetra Wallich. Attached and in the plankton of the Tar, Yadkin and Neuse Rivers, CP, P, (P).

PENNALES

Diatoma elongatum (Lyngb.) Agardh. Plankton, Cape Fear River, Columbus and Bladen counties, CP.

Actinella punctata F. W. Lewis. This species, long believed to be rare, is widespread in the Coastal Plain and abundant in winter, attached to plants in flowing water. In this country, it is now known from New Hampshire, New Jersey, and North and South Carolina.

In previous illustrations (Van Heurck, 1885; Boyer, 1916; Smith, 1950) the small spines along the edges of the frustule are either omitted or poorly delineated. These spines, however, are quite evident with proper preparation and illumination. Figures 10–11.

Pinnularia divergens W. Smith. In bottom ooze, Sleepy Creek, Wayne County, CP.

Pinnularia polyonca (Breb.) O. Muller. With the preceding species, CP.

Neidium apiculatum Reimer. In bottom ooze, Sleepy Creek, Wayne County, CP.

Neidium apiculatum var. *constrictum* Reimer. With the previous taxa, CP.

Stauroneis phoenicenteron Ehrenberg. In tow of Deep River, Randolph County, P. Previously reported from pond plankton in the Coastal Plain and mountains.

Frustulia rhomboides var. *saxonica* (Rabh.) de Toni. In bottom ooze, Sleepy Creek, Wayne County, CP.

Amphiprora paludosa W. Smith. In brown water creeks, Onslow County, CP.

Gomphonema acuminatum var. *coronata* (Ehr.) W. Smith. In small spring, Davidson County, P.

**Stenopterobia intermedia* (Lewis) Hustedt. Roadside ditch, east of Dover, Craven County, and bottom ooze from Sleepy Creek, Wayne County, CP.

Dinophyceae

PERIDINIALES

Peridinium limbatum (Stokes) Lemmermann. The second station for this rare species. In bottom ooze, Horse Pond, July, Carteret County, CP, (CP).

Cryptophyceae

CRYPTOMONADALES

Cryptomonas marsonii Skuja. (1948). Farm pond, Newton Grove, Sampson County, CP.

Myxophyceae

CHROOCOCCALES

**Eucapsis alpina* Clements and Shantz. On dead grass leaves with *Coleochaetae, Bulbochaete,* and diatoms in test pit, Yates Pond, April, 1960, Wake County, P.

Anacystis marina (Hansg.) Drouet and Daily. Along edge of Neuse River and on planted glass slides in Sleepy Creek, Halifax and Wayne counties, CP.

**Gloeochaete wittrockiana* Lagerheim. Among filamentous algae in seepage ditch in cool, shady water, south of Goldsboro, October, Wayne County, CP.

Entophysalis lemaniae (Ag.) Drouet and Daily. In rapids of creeks and Oconaluftee River, Swain, Wayne and Yadkin counties, CP, P, M.

Entophysalis rivularis (Kutz.) Drouet. In Sleepy Creek, Wayne County, CP, (P, M).

OSCILLATORIALES

Oscillatoria tenuis var. *natans* Gomont. Forming blue-green scum on rocks in current, Rocky Creek, July, 1959, Wake County, P.

Phormidium autumnale (Ag.) Gomont. In Boone Pond Swamp, Wake County, P, (P).

Anabaena spiroides var. *crassa* Lemmermann. Common in tow of Kerr Lake, Vance County, June, P.

**Wollea saccata* (Wolle) Bornet and Flahault. Floating gelatinous mass, Hester's Pond, Wake County, Aug. 11, 1960, CP.

Cylindrospermum licheniforme (Bory) Kutzing. On muddy banks of Neuse River, Halifax County, CP, (M).

**Leptochaete stagnalis* Hansgirg. On pebbles in creeks, June, Macon and Swain counties, M.

Rhodophyceae

NEMALIONALES

Batrachospermum globosporum Israels. In rapids, upper Little River, Harnett County, July, CP.

Batrachospermum sirodotii Skuja. On stones in shady rapids, Wake and Stanly counties, P. On wood and rocks in rapids of creek and Cullasaja River, Jackson and Macon counties, M. (CP, P).

Batrachospermum vagum (Roth) Agardh. Previously reported in a stream from the upper Coastal Plain (Whitford, 1943). This additional station is of interest because of the conditions of the water. The collection was made in Horse Pond, Carteret County, in three feet of water on sandy bottom. The water temperature at this depth was 32°C. and the *p*H was 6.0. CP.

LITERATURE CITED

BOURRELLY, P. 1957. Recherches sur les Chrysophycees. Rev. Algol. Mem. Hors-Sér. No. 1. 409 pp.

BOYER, C. S. 1916. The Diatomaceae of Philadelphia and Vicinity. Lippincott: Philadelphia.

DELPONTE, J. B. 1876. Specimen Desmidiacearum subalpinarum. Mem. R. Acad. Sci. Torino. 28.

IRENÉE-MARIE, F. 1938. Flore desmidiale de la région de Montreal. Laprairie, Canada.

JANE, F. W. 1940. Two new Chrysophycean flagellates: *Cyclonexis crinus* and *Synochromonas claeochrus*. Proc. Linn. Soc. 152 Sess. **40**: 298–309.

LUND, J. W. G. 1960. Some new or rare Chrysophyceae from the English Lake District. Hydrobiologia **16**: 97–108.

PASCHER, A. 1913–1927. Die süsswasserflora Deutschlands, Osterreichs und der Schweiz. Nos. 1–12. Jena.

———. 1939. Heterokonten. In L. Rabenhorst, Kryptogamen-Flora von Deutschland, Osterreich und der Schweiz. **11**: 1–1092.

PHILLIPS, R. C., AND L. A. WHITFORD. 1958. Additions to the fresh-water algae in North Carolina II. Castanea **23**: 25–30.

POCHMANN, A. 1957. Uber die kerbungen des zellrandes bei *Phacus* und eine ohnlicke bei einer Chrysomonad beobachtete erscheinung. Arch. Protistenk **102**: 44–83.

PRESCOTT, G. W. 1931. Iowa Algae. Univ. Iowa Studies Nat. Hist. 13. 235 pp.

———. 1951. Algae of the Western Great Lakes Area. Cranbrook Institute of Science, Bull. No. 31. Bloomfield Hills, Mich.

SCHUMACHER, G. J. 1958. An unusual algal habitat. Jour. Elisha Mitchell Sci. Soc. **74**: 141–142.

SCHUMACHER, G. J., AND L. A. WHITFORD. 1959. Additions to the fresh-water algae in North Carolina III. Jour Elisha Mitch. Sci. Soc. **75**: 29–32.

———, AND ———. 1959. Additions to the fresh-water algae in North Carolina IV. An annotated list of diatoms. Jour. Elisha Mitchell Sci. Soc. **75**: 101–106.

SKUJA, H. 1948. Taxonomie des Phytoplankton einiger Seen in Uppland Schweden. Symb. Bot. Upsal. 9. 398 pp.

SMITH, G. M. 1950. The Fresh-water Algae of the United States. McGraw-Hill: New York.

SMITH, R. C. 1958. Some additions to the fresh-water algal flora of North Carolina: algae occurring with Sphagnum. Jour. Elisha Mitchell Sci. Soc. **74**: 158–160.

STOKES, A. C. 1886. Notices of new fresh-water infusoria. Proc. Amer. Phil. Soc. **23**: 562–568.

VAN HEURCK, H. 1880–1885. Synopsis des diatomées de Belgique. Antwerp. 235 pp.

WHELDEN, R. M. 1939. Notes on New England Algae I. Cyclonexis and Actidesmium. Rhodora **41**: 133–137.

WHITFORD, L. A. 1943. The fresh-water algae of North Carolina. Jour. Elisha Mitchell Sci. Soc. **59**: 131–170.

———. 1956. Additions to the fresh-water algae in North Carolina I. Trans. Amer. Micros. Soc. **75**: 196–203.

———. 1958. Phytoplankton in North Carolina lakes and ponds. Jour. Elisha Mitchell Sci. Soc. **74**:143–157.

———. 1960. *Heterodictyon planctonicum* L. Whitford and *Chlorosaccus fluidus* Luther; further notes and corrections. Trans. Amer. Micros. Soc. **79**: 227–229.

———. 1960. Ecological Distribution of Fresh-water Algae. Pymatuning Symposia in Ecology, Sp. Pub. No. 2, Univ. of Pittsburgh, Pittsburgh, Pa.

HERPETOLOGICAL COLLECTIONS AND OBSERVATIONS IN HYDE AND TYRRELL COUNTIES, NORTH CAROLINA

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Introduction

This report is based upon joint herpetological collections and observations conducted in Hyde and Tyrrell counties, North Carolina, during the spring and summer of 1959 and 1960 and on one occasion in 1961. Collecting was centered in the area on the following dates: March 29; June 27, 28; July 28–31; and August 22, 1959; April 30; May 1; May 15; May 28, 29; June 27–29; July 1–3; and August 28, 1960. The single 1961 trip was made on May 20.

Although several workers have published data concerning various herpetological aspects

of the North Carolina Coastal Plain, there is but one available paper regarding significant collections from the northeastern portion: Brandt (1936) reported twenty-two species of frogs and toads from adjacent Beaufort County. Robertson and Tyson (1950) recorded a single specimen of *Sistrurus miliarius miliarius* from Hyde County (a snake which we observed to be relatively abundant), and Brimley (1944), in the latest work concerned with the herpetology of the state, mentioned but seven forms as being definitely recorded from the two counties; we failed to duplicate his records of *Alligator mississipiensis, Crotalus adamanteus,* and *Clemmys guttata.*

The present paper reports fifty-two species and subspecies: four salamanders, thirteen frogs and toads, six lizards, six turtles, and twenty-three snakes, all of which were actually collected or closely observed and identified beyond question by one or both authors. Small choruses of *Hyla ocularis* and *Hyla femoralis* were heard on the night of July 2, 1960; however, since no specimens of either species were collected or actually observed, both are omitted herein.

Undoubtedly transient collectors have secured specimens from the two counties and in turn have deposited these animals in various institutions throughout the country. We have made no attempt to locate such material or to review the literature for additional county records.

Both Hyde and Tyrrell counties are physiographically typical of the coastal plain region. The dominant hardwoods occurring over most of the area are sweet gum (*Liquidambar styraciflua*), southern red oak (*Quercus falcata*), swamp chestnut oak (*Q. prinus*), and red maple (*Acer rubrum*). Bottomland sections are dominated by bald cypress (*Taxodium distichum*), *Liquidambar,* and water and black tupelo (*Nyssa aquatica* and *sylvatica*). The loblolly (*Pinus taeda*) is the dominant pine, being replaced by the pond or pocosin pine (*P. serotina*) in many lowland tracts. Vast stretches of cane (*Arundiaria*), forming impressive cane brakes, are abundant in parts of Hyde County.

Lake Mattamuskeet, located in central Hyde County, is the largest fresh-water lake in the state and occupies some 50,000 acres inclusive of marginal swampland. New Lake, formerly known as Alligator Lake, is situated in the northwestern portion of the county and comprises approximately 12,500 acres. Hyde is bordered on the east and south by Pamlico Sound; on the west by the Pungo River, and on the north by Washington, Tyrrell, and Dare counties; the Alligator River separates Tyrrell and Dare. The intracoastal waterway bisects the county, joining the Pungo and Alligator rivers to the southeast and northeast; the latter river flows through northern Hyde and into New Lake.

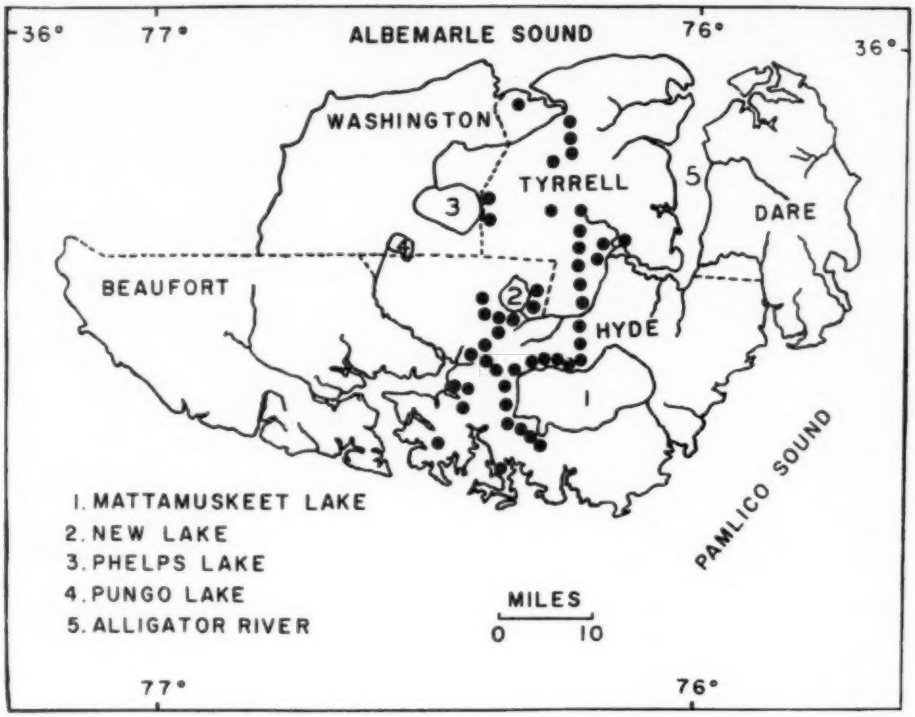
With the exception of Lake Phelps on its western boundry, Tyrrell County contains no large lakes, the main water courses being the Alligator and Scuppernong rivers on the east and north. Roadside canals, however, are quite numerous in both counties, and in some instances have resulted in the formation of small ponds.

Commercial lumber and pulp companies have invested in the area, providing logging roads through many otherwise impenetrable coastal forests. The aftermath of such operations has resulted in countless logs, slabs, and stumps in various stages of decomposition, as well as many deserted lumber camps and abandoned sawdust piles. Collecting in these habitats proved extremely productive, especially during the early spring months. Field activities through the warmer months of June, July, and August were conducted in the early morning and late afternoon, with attention concentrated upon the less-traveled paved roads from dusk until midnight.

We have departed from the nomenclature of Schmidt (1953) in only a few cases which conflict with that of Conant (1958). Common name usage conforms with those adopted by Conant and Committee (1956). Locality data were obtained from the mileage gauge of the speedometer and by the use of county maps procured through the State Highway Commission.

Specimens collected during the early part of this work are deposited in the North Carolina State Museum of Natural History (NCSM). Subsequent collections have been placed in the Field Collections of that institution (SMFC) and may be identified by accompanying numbers throughout this paper. Material recorded as DOR refers to examples seen dead on the road, which were not collected; specimens designated as AOR were collected alive on the road and are located in the collection unless otherwise stated.

We are indebted to the following people for their enthusiastic aid in the field and for financial assistance in transportation expenses: R. P. Rogers of Greenville, who accompanied us on all but one trip and who added substantially to our species list through his sedulous field work; M. J. Batchelor, Jr., of Wilson; E. D. Bruner, Jr., Robert B. Jackson, and C. L. Mandelin (United States Marine Corps), all of Raleigh; L. D. Dunnagan (United States Marine Corps) of Cherry Point; and Roger Rageot, Curator of Natural History at the Norfolk Museum, Norfolk, Virginia. We would further



MAP 1

thank F. B. Meacham and the Lake Mattamuskeet "Honker's Club," who kindly allowed our party to stay at their lodge during several excursions; John Worth Moore, for many courtesies and the opportunity to examine and record his growing collection of live Hyde County snakes; Austin B. Williams and Clarence E. Styron, University of North Carolina Institute of Fisheries Research at Morehead City, for help and criticism throughout the preparation of this paper; and B. B. Brandt, Professor Emeritus in the Zoology Department at North Carolina State College, and M. R. Carriker, Zoology Department at the University of North Carolina, for reading and criticizing the manuscript.

ANNOTATED LIST

Amphibia

Caudata

Ambystoma opacum Gravenhorst. Marbled Salamander. One specimen of this species (338) was collected beneath a rotting log on the east shore of New Lake, Hyde County, on June 29, 1959.

Amphiuma means means Garden. Two-toed Amphiuma. This aquatic amphibian is represented in the present survey by a single individual observed DOR thirteen miles north of Fairfield, Tyrrell County, on July 2, 1960. The example had probably attempted to cross the highway on the preceding night during heavy rainfall.

Desmognathus fuscus auriculatus Holbrook. Southern Dusky Salamander. One specimen, taken beneath a log at the edge of a temporary rain pool five miles east of Ponzer, Hyde County, on May 1, 1960, was typical of the subspecies. The characteristic dark venter and lateral "porthole" spotting were present and conspicuous. The individual, unfortunately, was lost during transportation.

Plethodon glutinosus glutinosus Green. Slimy Salamander. Two specimens of this common form (478–79) were secured under boards near an abandoned dwelling ten miles west of Swanquarter, Hyde County, on May 1, 1960. Another example, observed beneath a rotting log on the east shore of New Lake, Hyde County, on April 30, 1960, was not collected.

Salientia

Bufo quercicus Holbrook. Oak Toad. Ten specimens, representing individuals from both counties, are contained in the collection: (330) taken fourteen miles south of Columbia, Tyrrell County, on July 31, 1959; (324) collected under a log at the base of an evacuated sawdust pile four miles east, two miles south of Ponzer, Hyde County, on July 29, 1959; (476) secured by R. P. Rogers beneath a flat board in a canebrake thicket 8.5 miles east, six miles north of Ponzer, Hyde County, on May 1, 1960; (477) taken under a discarded sheet of tin near an abandoned lumber camp one mile west of Rose Bay, Hyde County, on May 1, 1960; (526–31) six specimens collected AOR along the New Lake road, Hyde County, by R. P. Rogers and the junior author on May 28, 1960.

This species was found to be extremely abundant following heavy rains, and under such conditions on May 28 many specimens were observed hopping across the dirt road from Highway 264 to New Lake. During the summer drought periods these toads were observed only rarely, and at such times invariably beneath some form of cover in moist situations.

Bufo terrestris Bonnaterre. Southern Toad. Four Hyde County specimens referred to this species were collected from the following two localities: (337) taken by R. P. Rogers beneath a rotting mattress eleven miles east of Belhaven on July 29, 1959; (635–37) three individuals collected under a slab pile along the east shore of New Lake on August 28, 1960, by Rogers.

Bufo woodhousei fowleri Hinekley. Fowler's Toad. Seven examples which we have designated as *fowleri* (638–41, 646–48) were secured by R. P. Rogers from slab piles along the east shore of New Lake, Hyde County, on August 28, 1960, in company with three specimens which we have previously called *terrestris*.

In the last two decades several investigators—Blair (1941), Gosner and Black (1956), and Volpe (1959)—have shown that *Bufo terrestris* and *B. w. fowleri* do hybridize in sympatric areas. Recently, Mrs. Norma Rothman (1960) informed us that hybridization apparently occurs among the larger *Bufo* inhabitants of the Albemarle–Pamlico Sound peninsula. Though inclined to agree with Mrs. Rothman, we consider our present collection insufficiently comprehensive to determine interspecific hybridization and have conducted no laboratory experiments in an effort to induce cross-breeding between the two entities. The eleven specimens at hand have been categorized temporarily as either *terrestris* or *fowleri* on the basis of cranial crest arrangements and dorsal and ventral spotting.

Acris gryllus gryllus Le Conte. Southern Cricket Frog. Six specimens of this common frog are contained in the collection: (551–56) collected from a flatwood pond 13.7 miles east of Ponzer, Hyde County, on May 29, 1960. We were unable to secure *gryllus* on the highways during nights of extreme precipitation; individuals were taken successfully only during the day in and about small ponds.

Hyla cinerea cinerea Schneider. Green Treefrog. Four Hyde County specimens are in the present collection: (373–75) three recently transformed juveniles taken by the junior author and C. L. Mandelin along the banks of a small pond at Ponzer on August 22, 1959; (539) collected behind a crate at the front of the Worth Moore Hobby Shop at Ponzer, on May 28, 1960, immediately following heavy rainfall. The owner tells us that the various forms of *Hyla* are commonly seen in large numbers about his shop during and at the end of heavy rain.

Hyla squirella Sonnini and Latreille. Squirrel Treefrog. Two examples of this very abundant *Hyla* were collected and preserved: (326) taken AOR twenty miles south of Columbia, Tyrrell County, on the night of July 30, 1959; (334) collected AOR three miles south of Columbia, Tyrrell County, on the evening of July 31, 1959. On these dates there were several severe thunderstorms in that area, and on both nights many *squirella* were observed along Highway 94 through an extensive swampy area between Fairfield, Hyde County, and Columbia, Tyrrell County.

Hyla versicolor versicolor Le Conte. Eastern Gray Treefrog. Three Tyrrell County specimens, collected AOR along Highway 94 on the night of May 28, 1960, are in the collection: (540) nineteen miles south of Columbia; (541–42) sixteen miles south of Columbia.

Pseudacris brimleyi Brandt and Walker. Brimley's Chorus Frog. A single specimen of this species is contained in the collection: (598) taken abroad on an overcast day along the New Lake road, Hyde County, on June 27, 1960, by R. P. Rogers.

The various members of *Pseudacris* are apparently abundant only during the early spring breeding congresses, appearing rarely at other times, probably with the advent of extreme rainfall.

Gastrophyrne carolinensis Holbrook. Eastern Narrow-mouthed Toad. Four specimens of this small anuran were collected from both counties; however, only two Tyrrell County individuals are preserved: (327) taken AOR twenty miles

south of Columbia on the night of July 30, 1959; (335) secured AOR three miles south of Columbia on the evening of July 31, 1959. A specimen taken under a rotting log two miles north of Swindell Fork, Hyde County, on June 27, 1959, was inadvertently lost. Another secured beneath a board along the shore of a small flatwood pond 13.7 miles east of Ponzer, Hyde County, on May 1, 1960, was not saved.

Rana catesbeiana Shaw. Bullfrog. Though apparently not uncommon, these large frogs are seldom seen except during and immediately following heavy precipitation. At such times they are frequently observed in the highways. Two specimens were collected under such conditions from Hyde County: an adult taken AOR on the night of July 30, 1959, four miles north of Fairfield was later released; another large specimen (Hampton Marine Museum, Morehead City, N. C., number 86) was collected AOR two miles north of Fairfield on the night of July 1, 1960, by Roger Rageot and the senior author.

Rana clamitans melanota Rafinesque. Green Frog. Four juvenile specimens (534–37) were collected on the night of May 28, 1960, at Kilkenny, Tyrrell County. A large *Natrix erythrogaster erythrogaster,* collected in Hyde County, regurgitated an adult *Rana* during capture. However, whether the frog was *melanota* or *catesbeiana* was not ascertained because of advanced digestive reactions.

Rana virgatipes Cope. Carpenter Frog. One specimen of this species was observed DOR along Highway 94, sixteen miles south of Columbia, Tyrrell County, on July 30, 1959. The species was heard in chorus near the site of the DOR on the same night, and again on the night of May 28, 1960, at both times from an area of inaccessible swamp.

Rana pipiens sphenocephala Cope. Southern Leopard Frog. This subspecies of *pipiens* was abundant along the roadside canals through both counties. Two specimens (518–19) were collected eleven miles east of Belhaven, Hyde County, from a temporary rain pool on May 15, 1960.

Reptilia

Chelonia

Chelydra serpentina serpentina Linnaeus. Common Snapping Turtle. Although represented in the present survey by a single specimen observed in a canal two miles west of Fairfield, Hyde County, on June 28, 1959, the form is probably common in all permanent bodies of water in both counties.

Sternotherus odoratus Latreille. Stinkpot. One example of this species was examined and later released; it was collected in a canal near Lake Phelps, at the Washington and Tyrrell County border on May 15, 1960.

Kinosternon subrubrum subrubrum Lacepede. Eastern Mud Turtle. Four specimens (502–05) are contained in the present collection, and were found in a canal 13.7 miles east of Ponzer, Hyde County, on the afternoon of May 15, 1960.

Terrapene carolina carolina Linnaeus. Carolina Box Turtle. Four individuals of this common chelonian were secured in Hyde County, and are presently in the collection of E. D. Bruner, Jr.: (2) AOR five miles north of Fairfield on June 28, 1959; (2) AOR along the New Lake road on the same date. Several others were seen DOR in both counties.

Chrysemys picta picta Schneider. Eastern Painted Turtle. One specimen of this colorful form was collected and is now in the collection of E. D. Bruner, Jr. The individual, an adult female, was collected AOR along the New Lake road, Hyde County, on June 28, 1959. Other examples were observed on logs in the canals bordering Highway 94 near Columbia, Tyrrell County.

Pseudemys scripta scripta Schoepff. Yellow-bellied Turtle. During the present work five specimens of this abundant race were taken by R. B. Jackson and the junior author from a roadside canal near Lake Mattamuskeet, four and one-half miles west of Fairfield, Hyde County, on May 15, 1960. These individuals are presently contained in the private collection of Mr. Jackson.

Sauria

Anolis carolinensis carolinensis Voigt. Carolina Anole. Two male specimens of this apparently uncommon lizard (469–70) were collected by R. P. Rogers on a rotting log along the east shore of New Lake, Hyde County, on May 1, 1960. These two preserved examples represent the only anoles seen during the survey.

Ophisaurus ventralis Linnaeus. Eastern Glass Lizard. The single specimen in the collection (328) was taken near an abandoned farm house ten miles west of Swanquarter, Hyde County, by R. P. Rogers on July 31, 1959. The individual is typical of the species in possessing no markings below the lateral groove.

Lygosoma laterale Say. Brown Skink. Though obviously quite common, the collection contains but one specimen (532) secured under a log on the east shore of New Lake, Hyde County, on May 28, 1960, by R. P. Rogers. Several other examples, collected in 1959 from

both counties, became desiccated and were not preserved.

Eumeces fasciatus Linnaeus. Five-lined Skink. This species is apparently the rarest of the genus in the area. Of many *Eumeces* collected and examined in the field, only two specimens possessed characters resembling those of *fasciatus*. One individual (311), a female, was taken guarding a clutch of ten eggs beneath a rotting log on the east shore of New Lake on June 28, 1959. Another specimen with nine eggs was discovered under the same log. This latter example eluded capture, however, and could not be specifically allocated. A female (557), containing five eggs, was secured on a rail fence along the east shore of New Lake on May 29, 1960. Both specimens are morphologically typical of piedmont *fasciatus*, having four supralabials anterior to the subocular and an enlarged middle row of subcaudals.

Eumeces laticeps Schneider. Broad-headed Skink. Two specimens of this large species are in the collection: (321), a female, 202 mm. over-all length, collected at the New Lake Lodge on the east shore of New Lake, Hyde County, on July 29, 1959, by R. P. Rogers; (506), an adult male, 279 mm. in total length, secured at the base of a pecan tree at Lake Phelps on the Washington and Tyrrell County border, on May, 15, 1960. The senior author (Palmer, 1959) reported a female of this species, in company with seventeen eggs, which was taken under a log two miles northwest of Swindell Fork, Hyde County, on June 27, 1959. This specimen, unfortunately, is no longer extant.

Eumeces inexpectatus Taylor. Southeastern Five-lined Skink. This species was without question the most abundant saurian throughout the two counties. Although but three specimens were actually collected and retained, at least fifty to seventy-five examples were caught, examined, and later released. The reduction in size of the middle row of subcaudals is pronounced, and the supralabials anterior to the suboculars almost always number four on each side. Abandoned sawdust piles and lumber camps provided ideal habitats for *inexpectatus;* as many as twenty or more individuals were observed in less than an hour at a single suitable site. All specimens encountered were secretive and were discovered only beneath logs, slabs, and similar cover. Locality data for the preserved material are as follows: (NCSM 59.3.28) an adult female, taken twenty miles south of Columbia, Tyrrell County, on March 29, 1959; (312) a male, collected five miles north of Fairfield, Hyde County, on June 28, 1959; (339) a juvenile, secured by R. P. Rogers at the New Lake Lodge along the east shore of New Lake, Hyde County, on July 29, 1959.

Serpentes

Natrix rigida Say. Glossy Water Snake. On the evening of May 20, 1961, a female specimen of this secretive water snake (782) was collected AOR two miles west of Fairfield, Hyde County. The snake measured 648 mm. total length immediately prior to preservation, and dissection of the hind-gut revealed the gastroliths of a crayfish.

Natrix erythrogaster erythrogaster Forster. Red-bellied Water Snake. Four adult specimens of this subspecies, representing material from both counties, were secured from the following localities: (303) a female collected AOR four miles north of Fairfield, Hyde County, in an area of extensive swamp on the afternoon of June 27, 1959; (495) a large female, 1219 mm. in length, taken in a canal 13.7 miles east of Ponzer, Hyde County, on May 15, 1960, which disgorged a large *Rana* during capture; (544) collected thirteen miles south of Columbia, Tyrrell County, on the night of May 28, 1960. A snake discovered in a temporary rain pool five miles east of Ponzer, Hyde County, on May 1, 1960, was sent alive to the San Diego Zoological Garden. DOR examples were commonly observed in both counties.

Natrix sipedon fasciata Linnaeus. Banded Water Snake. The present collection contains two specimens of this common snake: (496) a juvenile collected by R. P. Rogers beneath a flat board at the edge of a small flatwood pond, 13.7 miles east of Ponzer, Hyde County, on May 15, 1960; (543) an adult male taken AOR eighteen miles south of Columbia, Tyrrell County, on the night of May 28, 1960. Another adult, collected beneath a canal bridge three miles west of Fairfield, Hyde County, on June 28, 1959, became overheated and expired before it could be preserved. Several DOR specimens were observed in both counties, especially along Highway 94 between Fairfield and Columbia.

Natrix taxispilota Holbrook. Brown Water Snake. This large species is represented in the collection by a single specimen (318) collected in a goldfish pool at Ponzer, Hyde County, on July 28, 1959, by R. P. Rogers and the junior author. A small adult was seen DOR just south of Columbia, Tyrrell County, on July 3, 1960. Other live Hyde County specimens were observed in the Worth Moore collection at Ponzer.

Seminatrix pygaea paludis Dowling. Carolina Swamp Snake. The present specimen (331), an adult male, 356 mm. in total length,

was reported previously by the authors (1960). Collected AOR along Highway 94, fifteen miles south of Columbia, Tyrrell County, on the night of July 31, 1959, the snake represented a range extension of the subspecies. Dowling (1950) reported the previous northernmost record as Beaufort, Carteret County, some seventy miles to the south.

Storeria dekayi Holbrook. Brown Snake. The present collection contains eight specimens, seven of which are from Tyrrell County: (333) collected AOR fourteen miles south of Columbia on July 31, 1959; (377–78) taken AOR nineteen miles south of Columbia on August 22, 1959; (379) secured AOR two miles west of Gum Neck on August 22, 1959; (559) collected AOR sixteen miles south of Columbia on May 28, 1960; (599–600) taken under boards three miles south of Columbia on July 2, 1960; (603) collected by Roger Rageot beneath a rotting log on the east shore of New Lake, Hyde County, on July 2, 1960. Of the collected specimens, three were gravid females containing seven, ten, and eleven embryos.

We have elected the usage of the binomial in reference to this population since it seems apparent that the area is inhabited by a mixed assemblage of specimens exhibiting characters of intergradation between the subspecies *dekayi* and *wrightorum*. Earlier Trapido (1944) showed that the two races intergraded throughout most of North Carolina, and it was he who separated the two on the basis that *wrightorum* possesses a combined ventral and caudal scale total of 176 or more, as contrasted with 175 or less in *dekayi*. The former is reputed to exhibit a fused condition of the dorsal spotting as well (Trapido, *op. cit.*). In our animals the dorsal pattern is suggestive of *dekayi*, and a ventral-caudal computation of the sample results in a border-line arithmetic mean of 175.7.

Thamnophis sauritus sauritus Linnaeus. Eastern Ribbon Snake. Two Hyde County specimens of this race were collected and preserved: (NCSM 59.3.27) taken from a roadside canal at Fairfield on March 29, 1959; (381) collected on the Fairfield road, three miles north of Swindell Fork, on the night of August 22, 1959. A juvenile, discovered in the act of consuming a small toad, was taken under a log twelve miles north of Fairfield, Tyrrell County, on June 27, 1959. The snake later escaped from a collecting bag and was not recovered. Another immature individual was observed DOR five miles west of Gum Neck, Tyrrell County, on August 22, 1959.

Thamnophis sirtalis sirtalis Linnaeus. Eastern Garter Snake. Two adult specimens of this subspecies in which the typical longitudinally striped pattern was replaced by dark spotting were secured from Tyrrell County: (336) collected six miles west of Columbia on the night of July 31, 1959; (601) taken AOR ten miles south of Columbia on the evening of July 2, 1960, by L. D. Dunnagan and the senior author. Earlier, on March 29, 1959, a DOR specimen observed near Swindell Fork, Hyde County, also possessed a boldly spotted dorsum.

Haldea striatula Linnaeus. Rough Earth Snake. One specimen of this species (NCSM 59.3.25) was collected beneath a large rock in the yard of an abandoned dwelling on the east shore of New Lake, Hyde County, on March 29, 1959.

Heterodon platyrhinos Latreille. Eastern Hognose Snake. The specimen in the collection (594) is an adult female collected by R. P. Rogers in a slab pile along the east shore of New Lake, Hyde County, on June 27, 1960. A DOR example was seen along the New Lake road on May 28, 1960, and another was observed DOR on a logging road nineteen miles south of Columbia, Tyrrell County, on May 15, 1960.

Diadophis punctatus punctatus Linnaeus. Southern Ringneck Snake. The single specimen in the collection (465) is a female which was taken under a large board near an abandoned sawdust pile five miles east of Ponzer, Hyde County, on May 1, 1960. The snake is typical of the subspecies in possessing a broken neck ring, conspicuous labial spotting, a boldly marked venter, and a ventral-caudal count of 183 (143 ventrals, 40 caudals).

Carphophis amoenus amoenus Say. Eastern Worm Snake. Four specimens are contained in the collection: three (NCSM 59.3.26; SMFC 306, 461) were secured beneath logs and discarded tin on the east shore of New Lake, Hyde County; (380) collected two miles north of Fairfield, Hyde County, on the night of August 22, 1959.

Farancia abacura abacura Holbrook. Eastern Mud Snake. Three specimens of this abundant snake were collected: (545) taken along Highway 94 two miles west of Gum Neck, Tyrrell County, on the night of May 28, 1960; (605) secured AOR at the Northwest Fork Bridge, south of Columbia, Tyrrell County, on the evening of July 3, 1960, by L. D. Dunnagan. Another specimen, taken AOR five miles west of Fairfield, Hyde County, on the night of June 27, 1959, escaped in the laboratory and has not been recovered. Immediately following heavy rainfall many individuals were observed DOR in both counties, and all collected were

obtained on highways during or immediately following rain.

Coluber constrictor constrictor Linnaeus. Northern Black Racer. Two specimens of this common form were collected from Hyde County: (466) collected by Robert B. Jackson and the senior author eleven miles east of Belhaven on May 1, 1960; (467) secured eight and one-half miles east, six miles north of Ponzer at the site of a deserted sawdust pile on May 1, 1960. Numerous specimens were seen DOR in both counties, and several escaped capture in the field.

Opheodrys aestivus Linnaeus. Rough Green Snake. One specimen of *aestivus* is contained in the collection (308) from the yard of the New Lake Lodge on the east shore of New Lake, Hyde County, on June 28, 1959. Aside from this example, a juvenile was seen DOR eight miles north of Fairfield, Tyrrell County, on July 31, 1959.

Elaphe guttata guttata Linnaeus. Corn Snake. One subadult specimen of this colorful rat snake was taken from a small crevice at the top of an abandoned sawdust pile seventeen miles east of Ponzer, Hyde County, on May 1, 1960, and was later sent alive to the San Diego Zoological Garden. An adult is recorded from the Worth Moore collection, and another large example was observed DOR thirteen miles north of Fairfield, Tyrrell County, on July 2, 1960.

Elaphe obsoleta Say. Rat Snake. The present collection contains four specimens from Hyde County: (305) an adult male, 1321 mm. in total length, collected along the New Lake road on June 28, 1959; (457) a large male, 1829 mm. in length, secured from the Worth Moore Hobby Shop and originally collected two miles north of Ponzer; (472) an adult male, 1362 mm. in length, taken from a slab pile at an abandoned lumber camp eight and one-half miles east, six miles north of Ponzer by R. P. Rogers on May 1, 1960; (329) a small adult, collected by R. P. Rogers and the junior author inside a storage barn eleven miles west of Swanquarter on July 31, 1959.

The *Elaphe obsoleta obsoleta* x *E. o. quadrivittata* complex inhabiting the North Carolina Coastal Plain is a variable one which has led to much confusion regarding the status of the population. Conant (1958) referred to the typical Coastal North Carolina form as the "greenish rat snake" and considered it intermediate between *E. o. obsoleta* on the north and west and *E. o. quadrivittata* to the south. Brimley (1944), in discussing this lowland *Elaphe* and calling it *quadrivittatus* (=*quadrivittata*), was undoubtedly dealing with the greenish population, which becomes grayish or brownish in areas subject to *E. o. obsoleta* penetration. Robertson and Tyson (1950) reported typical *obsoleta*, with no variation toward the greenish phase, from Pitt County near Greenville, but further mentioned probable intergradation through much of the Coastal Plain.[1]

Since Hyde County is located in the northeastern Coastal Plain, genetic infiltration from *E. o. obsoleta* into the greenish population might be expected. In the available material all specimens resemble the greenish animal with the exception of (472), which appears somewhat intermediate. The venter of this specimen is characterized by prominent gray blotches which become more dense posteriorly. In life the dorsum is brownish with indistinct darker blotches; the four longitudinal stripes are not so conspicuous as in the other specimens. In addition, a snake seen DOR at the Washington and Tyrrell County border was very dark, and the dorsal and lateral stripes were almost obliterated. Two other dark-colored examples were observed in the Worth Moore collection.

We are once again conforming to the binomial pending examination of more material from the area and from other localities on the Coastal Plain.

Lampropeltis getulus getulus Linnaeus. Eastern Kingsnake. Six specimens of this abundant snake are in the collection from the New Lake area of Hyde County: (319) taken AOR by R. P. Rogers on July 29, 1959; (462) a juvenile secured beneath a flat board in the yard of an abandoned house on April 30, 1960; (464) collected by R. P. Rogers alongside a canal bordering the lake on May 1, 1960; (525) a subadult specimen taken by R. P. Rogers under a board at the site of a dismantled dwelling on May 28, 1960; (550) secured crawling abroad in a lowland thicket on May 29, 1960; (602) a small adult, collected AOR by L. D. Dunnagan on July 2, 1960. Aside from the examples contained in the collection, a large specimen was seen at the Worth Moore establishment, and a number of others were observed DOR in both counties.

Lampropeltis doliata doliata Linnaeus. Scarlet Kingsnake. One specimen of this secretive subspecies was taken from Hyde County: (463) an adult female, 559 mm. in total length, collected beneath a flat pine slab on the side of an abandoned sawdust pile one mile west of Rose Bay on April 30, 1960. The snake, ob-

[1] Through the courtesy of Mr. R. P. Rogers the SMFC contains several Pitt County *Elaphe obsoleta*, which are individually identifiable as the "greenish rat snake."

served in captivity for several months by the junior author, fed readily upon *Eumeces.*

Agkistrodon contortrix Linnaeus. Copperhead. Two large specimens of this species are in the collection: (661–62) taken under discarded pieces of tin on the east shore of New Lake, Hyde County, on August 28, 1960, by R. P. Rogers.

According to Gloyd and Conant (1943), the subspecies occurring through the area should be referable to the southern race, *contortrix.* However, Robertson and Tyson (1950) record apparent upland *mokeson,* or intergradient material, from contiguous Pitt County. Hyde County specimens from various institutions, examined by the senior author in conjunction with a study in the intergradation of the two races in North Carolina, possess somewhat intermediate characters. Therefore, we have postponed subspecific designation of this population.

Agkistrodon piscivorus piscivorus Lacepede. Eastern Cottonmouth. A single specimen (325) is in the collection from a canal six miles west of Swindell Fork, Hyde County, collected July 30, 1959. DOR examples were commonly seen in both counties, and several adults are recorded from the Worth Moore Hobby Shop.

Sistrurus miliarius miliarius Linnaeus. Carolina Pigmy Rattlesnake. Five Hyde County specimens are in the present collection from the following localities; (309) a juvenile taken at Ponzer on June 28, 1959, and received from the Worth Moore Hobby Shop; (320) an adult female, 559 mm. in total length, secured beneath a discarded sheet of tin along the New Lake road, eleven miles east of Belhaven on July 29, 1959; (323) an adult male, 584 mm. over-all length, collected by the junior author and R. P. Rogers from a dense pine-dominant flatwoods three and one-half miles east of Ponzer on July 30, 1959; (604) taken by L. D. Dunnagan and the senior author AOR at Scranton on the night of July 3, 1960, and presently a captive at the NCSM; (254) a large male, 597 mm. in total length, collected AOR along the New Lake road on August 28, 1960. Another specimen, taken AOR along the New Lake road on August 22, 1959, was sent alive to the Philadephia Zoological Garden.

It is interesting to note the erythristic dominant in these specimens, which is a marked contrast to the brownish and grayish examples occurring farther south on the Coastal Plain. All of our living animals were reddish and vividly patterned. The large size attained by the Hyde County population is also most interesting. Our largest specimen (254), a male, measured 597 mm. before fixation in formalin. Gloyd (1940), who studied available material from Alabama, Georgia, and North and South Carolina, reported a male taken at Bell Island, Hyde County, which measured 596 mm. as the largest specimen examined.

Crotalus horridus atricaudatus Latreille. Canebrake Rattlesnake. Four specimens of this large rattlesnake were collected from Hyde County: (310) a subadult, 762 mm. in length, taken on June 28, 1959, between Ponzer and New Lake; (322) an adult female, 1119 mm. in total length, collected at the edge of a canebrake thicket six miles north, eight miles east of Ponzer on July 29, 1959, by R. P. Rogers and the senior author; (474) a large adult, collected by members of a logging crew three miles east of Ponzer and received from the Worth Moore Hobby Shop, is presently a captive at the NCSM; (524) an adult female in the NCSM live collection, taken by R. P. Rogers and the junior author on May 28, 1960, beneath the edge of an abandoned dwelling along the New Lake road. A number of specimens were seen DOR in the Ponzer and Lake Mattamuskeet areas.

All individuals encountered were quite sluggish, becoming excited and defending themselves only after being molested or restrained. The specimen from the Ponzer cane brakes remained motionless in a compact coil while a slab pile less than a yard away was being investigated, and the snake was almost stepped on before being seen.

Despite the fact that several of the snakes examined possessed twenty-three scale rows at midbody, instead of the characteristic twenty-five as designated by Gloyd (1935) in his division of the monotypic *Crotalus horridus,* all pattern and color variations are applicable to *atricaudatus.*

LITERATURE CITED

BLAIR, ALBERT P. 1941. Variation, isolation mechanisms, and hybridization in certain toads. Genetics **26**: 398–417.

BRANDT, B. B. 1936. The frogs and toads of eastern North Carolina. Copeia **4**: 215–223.

BRIMLEY, C. S. 1944. Amphibians and reptiles of North Carolina. Reprinted from *Carolina Tips* (1939–43). Carolina Biological Supply Co.: Elon College, N. C. 63 pp.

CONANT, ROGER. 1958. A Field Guide to Reptiles and Amphibians. Houghton Mifflin Co.: Boston. 366 pp.

CONANT, ROGER, AND COMMITTEE. 1956. Common names for North American amphibians and reptiles. Copeia **1956**: 172–185.

DOWLING, HERNDON G. 1950. Studies of the Black Swamp Snake. *Seminatrix pygaea* (Cope), with

Descriptions of Two New Subspecies. Misc. Publ. No. 76, Mus. Zoology Univ. Mich. 38 pp.

GLOYD, HOWARD K. 1935. The cane-brake rattlesnake. Copeia **1935**: 175–178.

———. 1940. The Rattlesnakes, Genera *Sistrurus* and *Crotalus*. Spec. Publ. No. 4, Chicago Acad. Sci. 266 pp.

GLOYD, HOWARD K., AND ROGER CONANT. 1943. A synopsis of the American forms of *Agkistrodon* (copperheads and moccasins). Bull. Chicago Acad. Sci. **7** (2): 147–170.

GOSNER, KENNETH L., AND IRVING H. BLACK. 1956. Notes on amphibians from the upper Coastal Plain of North Carolina. Jour. Elisha Mitchell Sci. Soc. **72**: 40–47.

PALMER, W. M. 1959. Two large egg clutches of the broad-headed skink, *Eumeces laticeps*. Herpetologica **15**: 163.

PALMER, W. M., AND DAVID E. WHITEHEAD. 1960. A range extension of the swamp snake, *Seminatrix pygaea paludis* Dowling. Herpetologica **16**: 201.

ROBERTSON, WILLIAM B. AND EDWIN L. TYSON. 1950. Herpetological notes from eastern North Carolina. Jour. Elisha Mitchell Sci. Soc. **66**: 130–147.

ROTHMAN, NORMA. 1960. Information regarding *Bufo* hybridization in Hyde and Tyrrell counties, N. C. Personal communication.

SCHMIDT, KARL P. 1953. A Check List of North American Amphibians and Reptiles. Univ. Chicago Press: Chicago. 280 pp.

TRAPIDO, HAROLD. 1944. The snakes of the genus *Storeria*. Amer. Mid. Nat. **31**: 1–84.

VOLPE, E. PETER. 1959. Experimental and natural hybridization between *Bufo terrestris* and *Bufo fowleri*. Amer. Mid. Nat. **61**: 295–312.

ANOTHER HYPOTHESIS OF FUNCTION OF THE GLYCOGEN BODY OF BIRDS

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The morphology and embryogeny of the glycogen body and adjacent glycogen nuclei (lobes of Lachi and nuclei of Hofmann) have been thoroughly investigated by several authors (Gage, 1917; Olivo, 1926; Watterson, 1949, 1952; Frauchiger, 1952; Dickson and Millen, 1957). Other works have clarified numerous aspects of development, structure, and physiology of these bodies, but their functional place in the economy of birds has remained elusive. A number of hypotheses have been suggested, but none has proved to be operational; in all a nutrient role is assumed to exist, but in different processes: (1) embryogeny of the nervous system, or (2) development of tissues adjacent to the glycogen body, or (3) metabolism of body tissues in general.

Active participation of the glycogen body and its adjuncts in postembryonic events have been annoyingly difficult to prove since no investigator has yet been able to demonstrate an illuminating variation in glycogen content in response to any manipulated variable. Terni (1924) was unable to conclude whether the glycogen content was altered in unfed pigeons required to fly daily. Watterson *et al.* (1958), however, were confident that no reduction in glycogen content of the body occurred in 16-week chicks starved to death, although supportive data were not published. Szepsenwol (1953) could find in 6-week chicks no effect upon glycogen body, or its content, of vitamin-deficient diets, although the body did decrease or increase in size with protein-deficient and protein-rich diets respectively. These variations nevertheless suggest nothing of functional role of the body. In like category is the demonstration by Watterson *et al.* (1958) that hypophysectomy results in decrease in glycogen content of the body in embryo chicks.

It is conceivable, however, that if the glycogen body is indeed not altered by general inanition, it may by that fact be suspected of a role in neuronutrition, particularly if stress or inanition causes blood glucose levels to drop markedly (as appears to be the case), and if extraneural glycogen reserves can be demonstrated at the time to have become diminished. We have not here dealt with blood-sugar levels; Farner and his associates are investigating this aspect, and the literature is fairly extensive. As for extraneural glycogen, Van't Hoff (1909), Pratt (1941), and Streicher *et al.* (1950) have shown that the glycogen content of the liver is depleted with inanition in both chicks and pigeons. Nevertheless confirmation of Watterson and Doyle's unpublished data indicating that the glycogen body is unaffected by inanition is essential. Furthermore, although it may safely be assumed that if glycogen-body constancy actually does occur with inanition, it does so concurrently with depletion of liver glycogen, a

demonstration of this concurrence is of some importance.

Technique

Accordingly, we sought data bearing upon these two questions, utilizing 1- to 4-week chicks and adult pigeons of mixed sexes and undetermined age. The animals were starved for from 4 to 7 days and decapitated, and then weights and glycogen content of liver and glycogen body were obtained.

At once after decapitation of the birds, the livers were removed and homogenized in 95 per cent ethanol in a Waring blendor. About a 6 × 3 cm. section of the synsacrum containing the glycogen body was removed. The major portion of the tissues adhering to this section and the ventral wall of the vertebral column were removed before the synsacrum was placed in 95 per cent ethanol. Both the liver homogenate and the part of the synsacrum with the glycogen body were stored at 6°C. for 24 hours.

At this time the liver homogenate was washed several times with 95 per cent ethanol. The glycogen bodies were removed under a dissecting microscope and placed in tared centrifuge tubes. The liver homogenate and glycogen bodies were dried overnight in a vacuum desiccator using phosphorus pentoxide as a drying agent.

The tissues were weighed and powdered. Extraction of glycogen was carried out in the centrifuge tubes with perchloric acid (0.3 N) for 15 minutes in a boiling-water bath. The mixture was titrated to *p*H 7, using *p*H paper, with potassium hydroxide (1 N). The tubes were cooled under tap water and placed at 6°C. overnight.

These were then centrifuged. The supernate was analyzed for carbohydrate using a modification of the anthrone method described by Norris (1948). Some substance in the liver material interfered with the anthrone test. This was circumvented by a 24-hour precipitation of the perchloric acid-extracted supernate with 95 per cent ethanol at 6°C., washing the residue with absolute ethanol, absolute ethanol:ether (1:1), and ether. The precipitate was then suspended in water and centrifuged and the soluble portion in the supernate analyzed by the anthrone method.

The anthrone test was carried out by adding appropriate dilutions of the supernate to test tubes and adjusting this to 2 ml. with distilled water. The tubes were placed in an ice bath and 4 ml. of cold anthrone reagent (0.2 per cent and 95 per cent sulfuric acid) were added rapidly by syringe. The tubes were heated for 15 minutes in a boiling-water bath and then cooled in an ice bath. The tubes were read at 540 mμ in an Evelyn photoelectric colorimeter.

Glucose was used as a standard and the readings were converted to glycogen using the factor 0.90 (Norris, 1948).

Results

At the end of the experimental periods the weight of the animal, the weight of the liver, and the glycogen content of the liver were substantially decreased in birds that were starved (Table I), whereas there were no significant differences in the weight or glycogen content of the glycogen body in these same birds. There was an increase in weight and glycogen content of this body with increasing age in the chickens, as Szepsenwol and Michalski (1951) had previously shown. However, there was no close correlation between the body weight and the weight or the glycogen level of the glycogen body in 5-week-old chickens (Table II). Compared with glycogen in the liver the amount in the glycogen body was sizable in young chickens but small in older birds.

Birds starved until they died whose glycogen bodies were removed within 24 hours after death did not show a great decrease in the accumulation of glycogen in that structure (In a pigeon dead after 14 or 15 days of starvation, and two 1-week chicks dead after 3 or 4 days of starvation, the average weight of glycogen body was 7.1 and 7.5 mg. respectively, and the average weight of glycogen per body was 4.8 mg. in each).

Two factors probably made the quantities of liver glycogen detected lower than those actually present in that organ. First, trichloracetic acid extraction of chicken liver recovers only a fraction of the glycogen present, but in glycogen bodies all recoverable glycogen is extracted (Szepsenwol and Michalski, 1951). Perchloric acid extraction probably also recovers only a fraction of the liver glycogen. Second, the extra steps necessitated in the analysis of the livers undoubtedly substantially decreased the yield.

Even with this limitation, it is clear that the amount of glycogen in the glycogen body, if it goes down at all, does not go down as much as that in the liver upon starvation of the species tested. It is also evident that there is no correlation, increase or decrease, of the glycogen content of the glycogen body with variation in body weight in chickens of the same age. Finally, in an older bird the amount of glycogen in this organ obviously would be of little value in terms of supplying energy for the

Table I
Liver and glycogen-body analyses.

	Pigeons	Pigeons	1-wk. chicks	4-wk. chicks
Days duration	4.5	6.0	4.1	7.0
Starved				
Av. total wt./bird (gm.)	– (3)*	192 (6)	58 (4)	352 (6)
Av. wt. liver/bird (mg.)	740 (3)	725 (6)	265 (4)	1350 (6)
Av. glyc. content liver/bird (mg.)	– (3)	4.6 (6)	1.3 (4)	39.9 (6)
Av. wt. glyc. body (mg.)	6.7 (3)	6.5 (6)	8.2 (4)	14.6 (6)
Av. wt. glyc./glyc. body (mg.)	5.6 (3)	5.1 (6)	6.2 (4)	11.6 (6)
Fed				
Av. total wt./bird (gm.)	– (3)	288 (6)	126 (4)	573 (6)
Av. wt. liver/bird (mg.)	1135 (3)	1300 (6)	760 (4)	3000 (6)
Av. glyc. content liver/bird (mg.)	– (3)	110.3 (6)	8.4 (4)	142.1 (6)
Av. wt. glyc. body (mg.)	6.3 (3)	7.0 (6)	8.8 (3)	13.8 (5)
Av. wt. glyc./glyc. body (mg.)	4.7 (3)	5.3 (6)	6.1 (3)	11.2 (5)

* Parenthetical numbers indicate number of specimens in that sample.

Table II
Individual data for 4-week chick.

STARVED			FED		
Total bird wt. (gm.)	Glycogen body wt. (mg.)	Glycogen content per glycogen body (mg.)	Total bird wt. (gm.)	Glycogen body wt. (mg.)	Glycogen content per glycogen body (mg.)
320	9.9	8.4	650	—*	—*
335	14.8	9.9	575	12.5	10.6
260	15.2	12.1	550	9.1	7.5
330	18.4	14.7	455	19.4	15.4
455	18.2	14.6	525	12.7	10.1
410	12.7	9.6	680	15.1	12.6

* Missing due to laboratory accident.

entire body since total existence requirements of a 500 g. bird would be about 300 calories per day, whereas the yield of 10–15 mg. of glucose would be only .04–.06 calories.

Discussion

If, as is now apparent, the glycogen body does not decrease in size or content through inanition, even though bodily stores elsewhere, at least in the liver, are greatly depleted, it may safely be inferred that the stores of the glycogen body are not available even under severe stress for utilization at least outside the central nervous system. The location of the glycogen body below the meninges (Dickson and Millen, 1957) also argues against its having a function in general metabolism and favors a function closely related to the central nervous system.

The uniquely inefficient aspects of the circulatory system of the central nervous system, characteristic of all vertebrates, is well known. As a result of this inefficiency, under stress conditions the central nervous system is the first to succumb —not because of special qualitative needs of these tissues, but because of insufficient oxygen (the central nervous system utilizing 20 per cent of the total oxygen intake in a mammal) and insufficient nutrient supplies made available to cells, many of which even under normal conditions function within a narrow margin of safety. Brief and relatively small reduction in vascular content of these requirements surpass the margin of safety, and malfunction, unconsciousness, or death follow if the deficiencies are not promptly remedied. All other body cells operate within a much wider margin of safety. It has also been convincingly demonstrated that the energy requirements of the central nervous system are provided almost exclusively by metabolism of glucose; the energy

contributions of other substances are insignificant (Kety, 1957: 223-9).

Obviously any animal subjected frequently to such stress would have a strong selective advantage in possessing a reserve of oxygen or nutrients available specifically for the nervous system. Clearly birds, more than any other living vertebrates with the possible exception of bats, are frequently subjected to severe stress. In flight, especially over long distances and for great lengths of time, body reserves of glucose—the only nutrient metabolized by the nervous system—are greatly depleted, reaching almost nil in the liver. Such depletion would undoubtedly be reflected at least to a degree in blood-sugar level and thus render precarious the functioning of the central nervous system, since the margin of safety is already very narrow. Confirmation that blood-sugar levels indeed drop with stress is provided by Honeywell (1922), by Gulland and Peters (1930), and by the studies of Farner and his associates, although Riddle (1937) states that in the pigeon blood-sugar levels remain constant throughout life. Honeywell found a 6 per cent decrease after 48 hours of starvation in pigeons, and Gulland and Peters found a 11 per cent drop after 24 hours of starvation.

Although all birds are said to possess a glycogen body, and no other vertebrates do, not all existing birds—for example domestic chickens and flightless orders—meet flight-induced stress. Yet flight is a very basic characteristic of the the whole Class Aves, and other features employed in conjunction with the broad adaptive zone that class occupies have not been lost in "degenerate" types which have lost the ability of flight. If the glycogen body is important to the adaptive syndrome for flight, it would not be strange that the body should persist in flightless types just as do such other flight adaptations as hollow limb bones, air sacs, and flight feathers. Obviously such basic adaptations for flight as the keeled sternum have been lost in flightless birds, but the point is well taken that *some* structures adaptive to flight persist seemingly functionless in flightless birds. The glycogen body could reasonably fall into that category.

Therefore, it is our hypothesis that the glycogen body evolved in birds as a nutrient reserve for metabolic needs of the central nervous system under severe stress conditions, particularly of flight. It is not, therefore, regarded as functionally important in flightless types.

We envision that the processes of biosynthesis and utilization of glycogen in the glycogen body are similar to those that occur in liver or muscle (Fruton and Simmonds, 1958; Robbins, Traut, and Lipman, 1959). Glucose brought into the glycogen body by blood vessels is converted to glycogen. This is then released as glucose or glucosephosphate to the cerebrospinal fluid. Nerve tissues use these directly or store them in the form of glycogen for later use. This release carried out by the glycogen body is triggered by a stress-induced reduction of the blood-sugar level and parallel drop of the sugar level in the cerebrospinal fluid.

Under at least normal and possibly also under stress conditions, the glycogen in the glycogen body would be replaced continuously from its blood vascular system. Since the quantitative "margin-of-safety" needs of the central nervous system are minute, the glycogen content of the glycogen body would not be expected to decrease markedly when a bird is subjected to such a stress as starvation. The glycogen body obviously could not long provide *all* energy needs even of the CNS, since 15 mg. of glucose could provide total energy for a 5-g. CNS (which is about .01 of total body weight in an adult pigeon) for only about 50 minutes (extrapolated from data for man given by Kety, 1957). If our present hypothesis is correct, the role of glycogen could only be to elevate to safe levels the glucose content of cerebrospinal fluid that is receiving subminimal glucose from the blood due to the prolonged stress of sustained flight. Undoubtedly prompt replenishment of glucose in the glycogen body occurs whenever the stock in that body is drawn upon, but even if this were not true the glycogen body would be able to supply "marginal" needs —bringing subminimal quantities received by cerebrospinal fluid from the blood to survival levels—for at least several hours and perhaps as much as several days.

Conclusions

1. In starved adult pigeons and 1- to 4-week chicks the glycogen of the liver is almost depleted, whereas that of the glycogen body is not appreciably altered.

2. There is a marked increase in size and glycogen content of the glycogen body of 4-week as compared with 1-week chicks.

3. In birds of identical age, as indicated by experiments with 5-week chicks, there is no close correlation between weight of bird and weight or glycogen content of the glycogen body.

4. Proportion of glycogen-body weight to body weight diminishes greatly with increase in body size.

5. It is hypothesized that the glycogen body,

unique to birds, evolved as a nutrient reserve to provide a margin of safety for metabolic needs of the central nervous system when blood sources of glucose for cerebrospinal fluid become inadequate under the nearly unique severe stress of flight.

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LITERATURE CITED

DICKSON, A. D., AND J. W. MILLEN. 1957. The meningeal relationships of the glycogen body in the chick. Jour. Anat. **91**: 47–51.

FRAUCHIGER, E. 1952. Der Lumbalwulst (glycogen body) des Rückenmarkes der Vögel. Schweizer Archiv für Neurologie und Psychiatrie **69**: 400–402.

FRUTON, J. S., AND S. SIMMONDS. 1958. General Biochemistry. 2d ed. John Wiley: New York.

GAGE, S. H. 1917. Glycogen in the nervous system of vertebrates. Jour. Comp. Neurol. **27**: 451–65.

GULLAN, J. M., AND R. A. PETERS. 1930. Observations upon the reducing substances of pigeons' blood. Biochem. J. **24**: 91.

VAN 'T HOFF. 1909. Glycogen content of muscle of resting and flying pigeons. Sitzber. Ges. naturforsch. Freunde (through Biochem. Centr. **9**: 328).

HONEYWELL, H. E. 1922. Studies on the sugar in the blood of pigeons. Amer. J. Physiol. **58**: 152–167.

KETY, SEYMOUR S. 1957. The general metabolism of the brain *in vivo*. *In* Metabolism of the Nervous System, by Derek Richter, Sect. **6**: 221–237.

NORRIS, D. L. 1948. Quantitative determination of carbohydrates with Dreywood's anthrone reagent. Science **107**: 254–255.

OLIVO, O. 1926. Di alcuni caratteri fisici e chimici del corpo glicogenico del midollo lombo-sacrale degli uccelli. Boll. Soc. Biol. Sperimentale **1**: 81–84.

PRATT, IVAN. 1941. The effect of *Emeria tenella* (Coccidia) on the glycogen stores of chickens. Am. J. Hyg. **34** C: 54–61.

RIDDLE, O. 1937. On carbohydrate metabolism in pigeons. Cold Spring Harbor Symposia Quant. Biol. **5**: 362.

ROBBINS, R. W., R. R. TRAUT, AND F. LIPMANN. 1959. Glycogen synthesis from glucose, glucose 6-phosphate, and uridine diphosphate glucose in muscle preparations. Proc. Amer. Acad. Sci. **45**: 6–12.

STREICHER, E., D. B. HACHEL, AND W. FLEISCHMAN. 1950. Effects of extreme cold on the fasting pigeon with a note on survival of fasting ducks at –40°C. Am. J. Physiol. **161**: 300.

SZEPSENWOL, J. 1953. Effect of various diets on the glycogen body of the chick. Fed. Proc. **12**: 141.

SZEPSENWOL, J., AND J. V. MICHALSKI. 1951. Glycogenolysis in the liver and glycogen body of the chicken after death. Am. J. Physiol. **165**: 624–7.

TERNI, J. 1924. Ricerche sulla cosidetta sostanza gelatinosa (corpo glicogenico) del midollo lombosacrale degli uccelli. Arch. ital. anat. e embriol. **21**: 55–86.

WATTERSON, R. L. 1949. Development of the glycogen body of the chick spinal cord I. Normal morphogenesis, vasculogenesis and anatomical relationships. J. Morph. **85**: 337–390.

———. 1952. Development of the glycogen body of the chick spinal cord III. The paired primordia as revealed by glycogen-specific stains. Anat. Rec. **113**: 39–52.

WATTERSON, R. L., P. VENEZIANO, AND D. A. BROWN. 1958. Development of the glycogen body of the chick spinal cord V. Effects of hypophysectomy on its glycogen content. Physiol. Zool. **31**: 49–59.

SOME AQUATIC PHYCOMYCETES FROM LAKE TEXOMA[1]

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The latter part of the summer of 1960 was spent at the University of Oklahoma Biological Station at Willis, Marshall County, Oklahoma, surveying the aquatic Phycomycetes of Lake Texoma (formed by impoundment of the Red and Washita rivers in 1942 and bounded by Oklahoma and Texas). This initial investigation involved morphological and taxonomic studies of some phycomycetous parasites and saprophytes of algae, or other fungi, and of pollen grains.

Lake-water collections were made from the

[1] Supported by a National Science Foundation grant-in-aid administered by the University of Oklahoma Biological Station and a grant from the Texas A & M College Fund for Organized Research.

shore line of well-protected inlets where the water depth was usually from two inches to one foot. Sites especially sought were those in which algae abounded, for these plants harbor phycomycetous parasites and saprophytes. The water collections were returned to the laboratory, placed in separate petri dishes, and "baited" with pollen of sweet gum (*Liquidambar styraciflua* L.) and pine (*Pinus taeda* L.). After two to three days the pollen bait was examined for fungi. During this period of incubation, the algae in the collections were surveyed for epiphytic and endophytic parasites.

Water collections were taken from various coves and inlets of Buncombe Creek, from along the shore from the Biological Station southwest to a willow swamp located approximately one-fourth mile east of the Willis Bridge Site, from the mouth of Cowan Creek (a spring-fed stream) just west of the Willis Bridge Site, and from small ponds on Island Number Two, which is located southeast of the Biological Station. Nineteen collections were made from the Blue River at the Oklahoma State Highway 7 bridge, Johnston County.

The following is a description of the phycomycetous fungi collected. The isolates listed below represent the first report of these species or of any Phycomycetes from the Lake Texoma area; all are also new to the State of Oklahoma. The fungi are listed following the systematic account as given by Sparrow (1960).

Chytridiales

Olpidium entophytum (Braun) Rabenhorst parasitic in *Spirogyra* sp., collected one-half mile east of Willis Bridge Site, August 14, 1960.

Olpidium pendulum Zopf in sweet-gum pollen bait, from shallow water in far inlet located between the University of Oklahoma Biological Station and Arrow Head Point, August 24, 1960.

Phlyctidium megastomum Sparrow on sweet-gum pollen bait, collected from far inlet between the Biological Station and Arrow Head Point and from the first large inlet in Buncombe Creek, one-half mile from the Station, August 25 and August 4, 1960, respectively (Figs. 9a–e, 17, 18).

This species is based on a chytrid misidentified by Raitschenko (1902; Sparrow, 1943). Raitschenko described this as a parasite of *Anabaena flos-aquae;* he indicated that the fungus also can live saprophytically, but did not name the substrate.

The sporangia of the Texoma isolate were not as large (15–40 μ in diameter) as those described by Raitschenko, which reached a diameter of 70 μ. The endobiotic portion of the thallus had the form of a short peg, distally expanded to form a large spherical (5–14 μ in diameter) or ellipsoidal (8–15 μ long by 8–13 μ wide) unbranched swelling. During the present study, even with the use of clearing agents, potassium hydroxide and lactophenol, no rhizoids were seen on the endobiotic thallus of this fungus.

Rhizophydium sphaerotheca Zopf on sweet-gum and pine pollen baits, from Willow Swamp Inlet, August 4, 1960. The size of sporangia (14–33 μ in diameter) reported is somewhat larger than usually seen in this species. Perhaps this isolate is similar to the multiporous form called *R. pollinis-pini* by Zopf wherein sporangia were reported up to 36 μ in diameter (Sparrow, 1960).

Rhizophydium pollinis-pini (Braun) Zopf on sweet-gum and pine pollen baits, Buncombe Creek, August 5, 1960 (Figs. 2a–e, 13). This present isolate had a branched rhizoidal system similar to that previously described for the species. However, in unifungal cultures, I found one apophysate thallus on the same pollen grain with another nonapophysate thallus (Fig. 13). This condition points up again the questionable taxonomic value of the apophysis in separating the genera *Rhizophydium* and *Phlyctochytrium*. This problem has previously been noted in other species of these genera by Couch (1932) and Koch (1957).

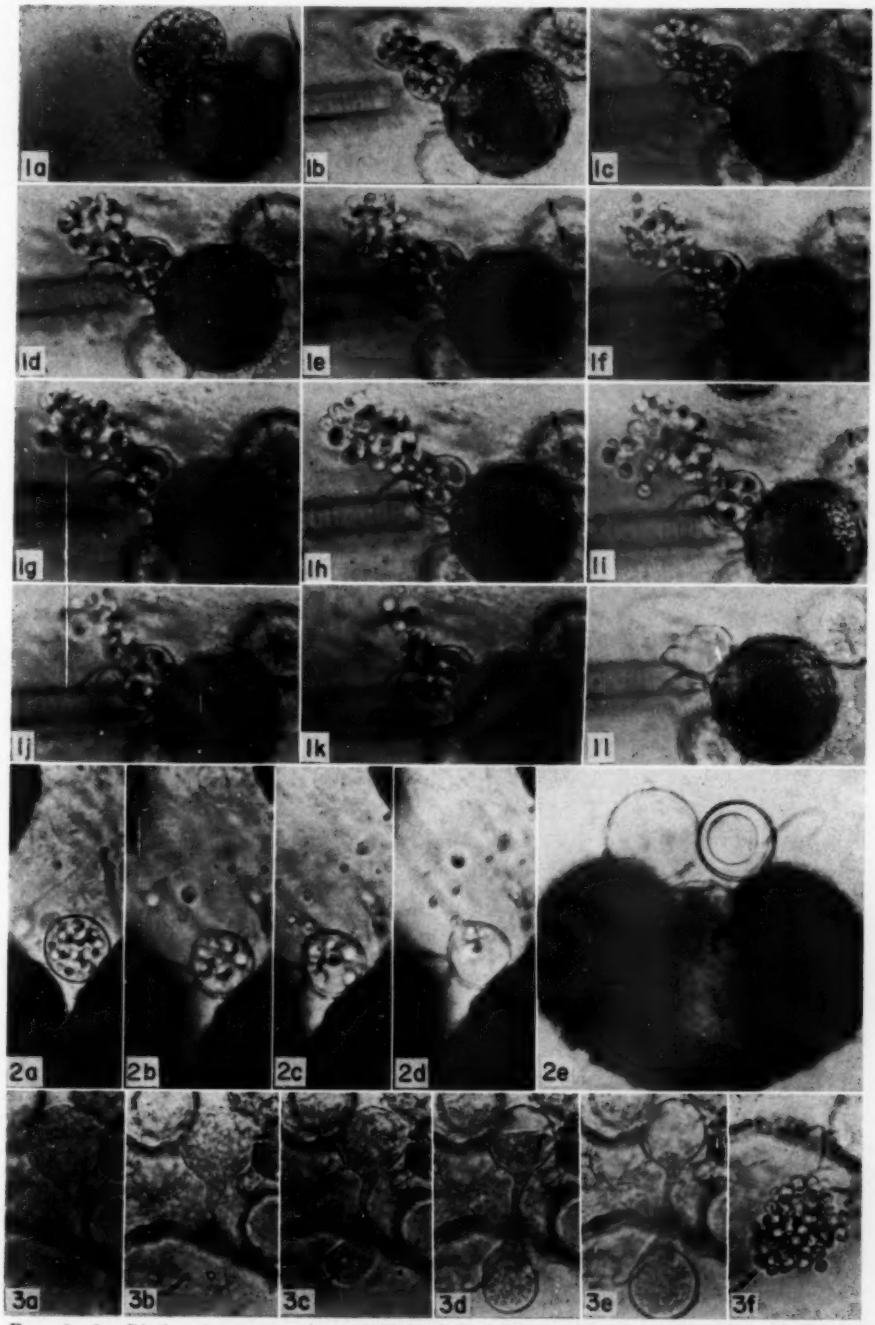
Rhizophydium minutum Atkinson parasitic on *Spirogyra* sp. from Willow Swamp Inlet, July 30, 1960 (Figs. 19, 20, 21). This is the second record of the species in the United States.

Phlyctochytrium irregulare Koch parasitic on *Spirogyra* sp. and on sweet-gum pollen bait, from several sites along the lake shore from the Biological Station to Willow Swamp Inlet, July 31, 1960 (Figs. 4a–e, 16, 22).

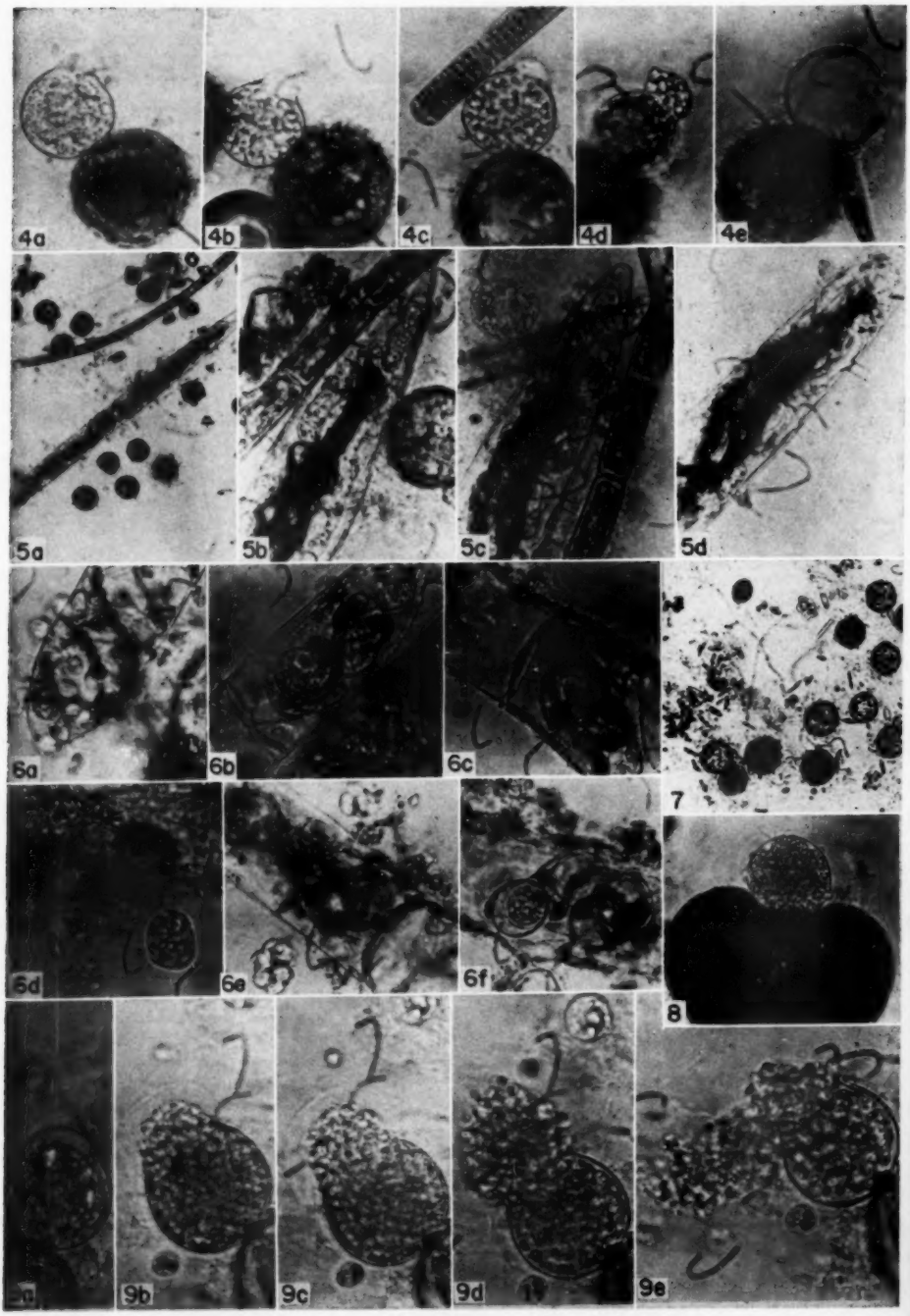
This is the third report of this species from the United States and the first as a parasite of *Spirogyra*. As pointed out by Koch (1957) the sporangial phase of *P. irregulare* is very similar to that of *P. hallii* Couch (1932), a parasite of *Spirogyra*. However the wall of the resting cell of *P. hallii*, unlike the smooth-walled resting spore of *P. irregulare*, has blunt spines. No spiny resting bodies were seen in this material.

Phlyctochytrium palustre Gaertner on sweet-gum and pine pollen baits, Buncombe Creek, August 6, 1960 (Figs. 8, 14, 15).

This is the second record of its occurrence and the first in the United States. It is very difficult to identify to species several of the multiporous phlyctochytriums. There is considerable variability in size and shape of sporangia and apophyses, in number and length of discharge papillae, and in rhizoidal arrange-



FIGS. 1a–1. Discharging sporangium of *Chytridium lagenaria* on sweet-gum pollen. X 574. FIGS. 2a–d. Discharging sporangium of *Rhizophydium pollinis-pini* on pine pollen. X 630. FIG. 2e. Resting spore (right) of *R. pollinis-pini*. X 630. FIGS. 3a–f. Discharging sporangium of *Rhizidiomyces hansonii* on sweet-gum pollen. X 382.



FIGS. 4a–e. Development and discharge of sporangium of *Phlyctochytrium irregulare* on sweet-gum pollen. X 511. FIGS. 5a–d. *Lagenidium rabenhorstii* in *Closterium* sp. FIG 5a. Single moribund *Closterium*

ments. At present several multiporous species are separated mainly on the basis of substrate differences.

The Texoma isolate seems to fit best the designation *P. palustre*. The sporangia, measuring 10–31 μ in diameter, are spherical, angular (rarely), or truncate when only two oppositely placed apical papillae are present. Two to eight (usually three to five) gelatinous papillae (3–4 μ long by 3 μ at the base) may be present. The apophysis may be spherical or the main rhizoidal axis may be slightly expanded; branched rhizoids are present.

Chytridium lagenaria Schenk on sweet-gum pollen bait from Willow Swamp Inlet and Blue River at Oklahoma Highway 7, July 31 and August 21, 1960, respectively (Figs. 1a–l, 10, 11).

C. lagenaria has been prevoiusly reported as a parasite and saprophyte of algae. The present isolate is larger than other apophysate members of the genus; this is the case with *C. inflatum* Sparrow (Sparrow, 1933) which ". . . is doubtfully separate from *C. lagenaria*. . . ." (Sparrow, 1960). It differs from *C. citriforme* Sparrow (1952) in not being citriform and lacking a subapical papilla, which gives the latter a tilted aspect. This present isolate also lacks the conical umbo operculum of *C. brevipes* Braun; it is considerably larger in all aspects than *C. scherffelii* (Scherffel) Sparrow. Since it falls within the species concept of *C. lagenaria*, it seemed best not to erect a new species merely because it grows on sweet-gum pollen.

Hyphochytriales

Rhizidiomyces hansonii Karling on sweet-gum pollen bait and less frequently on pine pollen bait, Buncombe Creek, August 9, 1960 (Figs. 3a–f, 12).

This is the first report of this fungus since its discovery in Brazil (Karling, 1944). In the present isolate the sporangia were occasionally found on a stout extramatrical nonapophysate rhizoid (Fig. 12).

Nine isolates of the organism were purified and grown on agar media from zoospores to sporangia, which in turn discharged. All isolates lived and multiplied for three days, after which time growth stopped.

Saprolegniales

Aphanomycopsis bacillariacearum Scherffel in sweet-gum pollen bait from Blue River water samples taken at Oklahoma State Highway 7, August 21, 1960 (Fig. 7).

This is the first report of the occurrence of this genus on pollen. Previously it has been reported as a parasite of some diatoms and desmids. Only a sparse amount of material was available for study and resting spores were not seen. Consequently its identification should be considered questionable. On the other hand, the holocarpic endobiotic branched tubular thallus is characteristic of only this genus in the Ectrogellaceae and is not characteristic of *Aphanomyces* de Bary or *Plectospira* Dreschsler (1927), with which it could conceivably be confused on the basis of spore discharge.

It seems to fit best the description of *A. bacillariacearum*, although, like *A. desmidiella* Canter (1949), it lacks the *spriezapparat*. Perhaps the lack of this so-called spreading apparatus, which apparently aids in forcing diatom valves apart, is merely a reaction to such a host, there being no special penetrating or spreading problem in desmids or pollen grains.

Lagenidiales

Olpidiopsis luxurians Barrett, parasitic in *Aphanomyces* sp. growing on snake-skin and cow-horn bait, from water samples taken a few hundred yards west of the Biological Station, July 21, 1960.

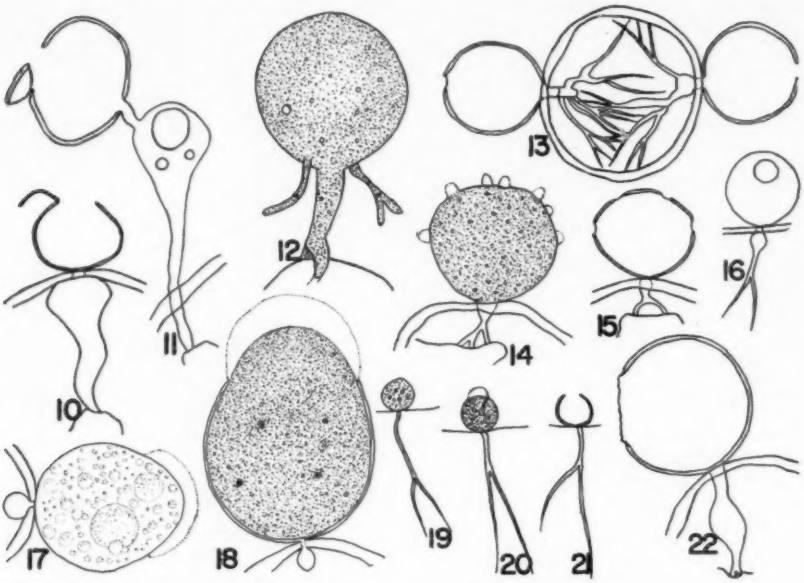
Myzocytium proliferum Schenk in *Spirogyra* sp., from Blue River at Oklahoma Highway 7, August 17, 1960 (Figs. 6a–f).

Lagenidium pygmaeum Zopf in sweet-gum pollen bait, from a small inlet a few hundred yards west of the Biological Station, July 21, 1960.

Lagenidium rabenhorstii Zopf in moribund *Closterium* sp. collected from a far inlet located between the Biological Station and Arrow Head Point, August 24, 1960 (Figs. 5a–d).

This isolate, while parasitizing a species of *Closterium*, differs from *L. closterii* de Wildeman (1893) by lacking a spherical or subspherical structure just beneath the point of constriction of the discharge tube, as it passes through the host cell wall, and by having a less

with parasite discharge tubes extending to outside. X 100. FIG. 5b. Tubular thallus of parasite. X 450. FIGS. 5c, d. Mature and empty sporangia of parasite. X 450. FIGS. 6a–f. *Myzocitium proliferum* in *Spirogyra* sp. X 472. FIG. 6a. Zoospores penetrating host cell. FIGS. 6b, c. Zoosporangia and discharge tube. FIGS. 6d, e. Discharging zoosporangia. FIG. 6f. Empty male gametangia and female gametangium with resting spore. FIG. 7. *Aphanomycopsis bacillariacearum* in sweet-gum pollen. Discharge tube with encysted zoospores. X 268. FIG. 8. Sporangium of *Phlycotochytrium palustre* on pine pollen. X 420. FIGS. 9a–e. Young sporangium and sporangial discharge of *Phlyctidium megastomum* on sweet-gum pollen. X 574.



FIGS. 10–22. All from sweet-gum pollen except where otherwise indicated. X 700. FIGS. 10, 11. Empty sporangia of *Chytridium lagenaria*. Note extensive extramatrical development shown in Fig. 11. FIG. 12. Sporangium of *Rhizidiomyces hansonii*. FIG. 13. Apophysate (right) and nonapophysate (left) isolate of *Rhizophydium pollinis-pini*. FIGS. 14, 15. Developing and discharged sporangia of *Phlyctochytrium palustre*. FIGS. 16, 22. Empty sporangia of *P. irregulare*. FIG. 16 from *Spirogyra*. FIGS. 17, 18. Sporangia of *Phlyctidium megastomum*. FIGS. 19–21. *Rhizophydium minutum* on *Spirogyra*.

extensive thallus. The present fungus approaches the description of the imperfectly known *L. intermedium* de Wildeman (1895), also a parasite of *Closterium;* however, it is dissimilar to the illustrations of the latter.

ACKNOWLEDGMENTS

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LITERATURE CITED

CANTER, H. M. 1949. On *Aphanomycopsis bacillariacearum* Scherffel, *A. desmidiella* N. Sp., and *Ancylistes* spp. in Great Britain. Trans. Brit. Mycol. Soc. **32**: 162–170.

COUCH, J. N. 1932. *Rhizophidium, Phlyctochytrium,* and *Phlyctidium* in the United States. Jour. Elisha Mitchell Sci. Soc. **47**: 245–260.

DRECHSLER, C. 1927. Two water molds causing tomato rootlet injury. Jour. Agr. Res. **34**: 287–296.

KARLING, J. S. 1944. Brazilian anisochytrids. Amer. Jour. Botany. **31**: 391–397.

KOCH, W. J. 1957. Two new chytrids in pure culture, *Phlyctochytrium punctatum* and *Phlyctochytrium irregulare*. Jour. Elisha Mitchell Sci. Soc. **73**: 108–122.

RAITSCHENKO, A. A. 1902. Ueber eine Chytrideacee: *R. sphaerocarpum* (Zopf) Fischer. Bull. Jardin Impér. Bot. St. Petersb. **2**: 124.

SPARROW, F. K., JR. 1933. Observations on operculate chytridiaceous fungi collected in the vicinity of Ithaca, N. Y. Amer. Jour. Botany **20**: 63–77.

———. 1943. The Aquatic Phycomycetes, Exclusive of the Saprolegniaceae and *Pythium*. University of Michigan Press: Ann Arbor.

———. 1952. A contribution to our knowledge of the Phycomycetes of Cuba. Rev. Soc. Cubana Bot. **9**: 1–19.

———. 1952. Aquatic Phycomycetes. University of Michigan Press: Ann Arbor.

WILDEMAN, É. DE. 1893. Notes mycologiques II. Ann. Soc. Belge Micro. (Mém.) **17**: 35–63.

———. 1895. Notes mycologiques IV. Ann. Soc. Belge Micro. (Mém.) **19**: 59–80.

DEHISCENCE OF APOTHECIA OF THE CUP FUNGUS *PATELLA ALBIDA* (SCHAEFF.) SEAVER

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Introduction

Corner (1929) established that there are two general types of apothecial development among cup fungi—gymnocarpic and angiocarpic. In the latter kind, a cortical layer together with inward growth of the marginal hyphal tissue forms a sheathlike covering over the developing ascogonia from which the hymenium arises. Eventually pressure from the expanding hymenium causes the rupture of the covering layer and thus terminates in exposure of the hymenium. Such a covering is wanting, however, among gymnocarpic species of cup fungi.

The writer's interest in the mechanism of opening of angiocarpic cup fungi was stimulated by the findings of Seaver (1937) from studies of *Urnula geaster* Peck. Seaver noted that pressure for the opening, splitting, and expansion of the cigar-shaped apothecia and for explusion of the ascospores is caused by the paraphyses. During these progressive developmental processes the paraphyses become swollen and are transformed thereby from filamentous structures into series of inflated, beadlike or monilioid, catenulate cells. Subsequently it was found (Wolf, 1958) that a similar transformation involves the paraphyses of *Urnula craterium* (Schw.) Fries, but it is confined to a narrow zone at the rim of the cup. The change in shape and increase in size of the cells composing the paraphyses of *U. craterium* were stated to account for apical fissuring of the apothecia during initial stages of dehiscence and for expulsion of the ascospores from asci in the region near the apothecial rim. Glycogen was hydrolyzed to reducing sugars in the tissues near the rim, *i.e.* in the asci and paraphyses, thus increasing the osmotic pressure with resultant swelling of the cells composing the paraphyses.

So far as can be determined little consideration has been given to mechanisms involved in apothecial opening among other genera of angiocarpic cup fungi. The following account deals with such a study of one of the Pezizaceae, *Patella albida*.

Observations

During the current year numerous apothecia of *Patella albida* were noted to be growing on soil beneath the benches in greenhouses at Duke University. It is well known that this organism occurs widely in eastern North America and was first collected in Europe approximately 200 years ago. Its apothecia are sessile and cupulate, they attain a diameter of from 2 to 3 cm., and they possess on the outer surface a coating of stiff, septate, pointed, brown setae. The ascospores measure 25–27 X 12–15 μ and are ellipsoid with a minutely papillate wall, and characteristically each spore has two prominent oil globules. To date all efforts to germinate these ascospores in water or on potato dextrose agar have resulted in failure.

Abundant apothecia in various stages of development were available for morphologic studies, using free-hand sections and appropriately fixed and stained preparations, embedded in paraffin. Young apothecia are almost spherical, being slightly flattened or indented above. At this early stage the cells composing the rim periphery are markedly larger than those of the hypothecium, and bulbous cells are thickly interspersed among the setae around the margin of the rim. These bulbous cells are of two kinds —thin-walled, hyaline ones and thick-walled ones. Presumably the latter kind arise from the former. At first the bulbous cells tend to be

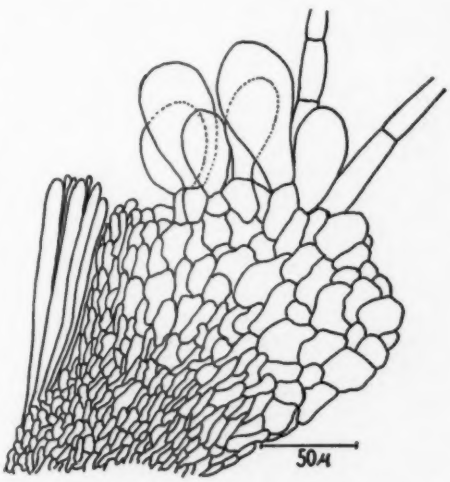

FIG. 1. Vertical section of tissues composing the apothecial rim of *Patella albida*.

inwardly directed, but as the cups open they stand vertically, (Fig. 1) as shown by examination of more mature cups.

Initiation of opening of the apothecia of *P. albida* is herein interpreted to be caused by expansion of the bulbous cells that surround the orifice of the cup, together with the expansion of the cells that compose the tissues of the rim. Subsequently all further expansion, ending when the cup is fully mature, is probably a response to increase in diameter of the developing asci.

Whether the dehiscence of apothecia of other Pezizaceae is caused by similar structures and their expansion during growth is not known.

Summary

Special bulbous cells are formed on the apothecial rim of *Patella albida* that are interpreted to function in initiation of apothecial dehiscence. Early expansion of apothecia is, additionally, a response to increase in size of cells composing the tissues of the rim. Subsequent apothecial expansion is caused by increase in diameter of the asci.

LITERATURE CITED

CORNER, E. J. H. 1929. Studies in the morphology of Discomycetes. II. The structure and development of the ascocarp. Trans. British Mycol. Soc. **14**: 275–291.

SEAVER, F. J. 1937. Photographs and descriptions of cup fungi XV. Urnula geaster. Mycologia **29**: 60–65.

WOLF, F. A. 1958. Mechanism of apothecial opening and ascospore expulsion by the cup fungus Urnula craterium. Mycologia **50**: 837–843.

STUDIES ON THE IMMUNITY RELATIONSHIPS IN WHITE MICE GIVEN INFECTIONS WITH *NEMATOSPIROIDES DUBIUS* BAYLIS, 1926 (NEMATODA: HELIGMOSOMIDAE)[1]

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Ability to acquire immunity to reinfection by parasitic helminths has been demonstrated using an extensive list of examples: *Trichinella spiralis* (McCoy, 1931), *Strongyloides ratti* (Sheldon, 1937), *Nippostrongylus muris* (Africa, 1931; Schwartz *et al.*, 1931) *Ascaris lumbricoides* (Kerr, 1938) *Ancylostoma caninum* (Otto and Kerr, 1939), *Haemonchus contortus* (Stoll, 1929), and *Trichostrongylus calcaratus* (Sarles, 1932). In the case of *Nematospiroides dubius*, available reports indicate that attempts to produce such immunity have been unsuccessful (Spurlock, 1943; Baker, 1955; Liu and Ivey, 1958). Perhaps the failures were due to the use of only one stimulating infection given prior to the challenging infection, which, as in certain other cases, might produce an immunity too feeble to be demonstrated by the methods used. In any event, it seemed worthwhile to carry out the present tests in an attempt to demonstrate acquired immunity in mice by giving repeated infections with this parasite.

[1] A portion of a dissertation submitted to the Faculty of the University of North Carolina in partial fulfillment of the requirement for the degree of Doctor of Philosophy in the School of Public Health.

Materials and Methods

A strain of Swiss white mice maintained in this department for a number of years was used in this study. Equal numbers of males and females 2.5 months old at the beginning of each experiment were used.

The life cycle of *N. dubius* has been described by Ehrenford (1954). Eggs are passed in the feces of the host and hatch in approximately 26 hours at temperatures of 23–26°C. (provided the fecal material has been kept moist). The free-living larvae require from 4 to 6 days to become infective and entry into the host is by ingestion. Within from 24 to 48 hours after ingestion, the larvae penetrate the mucosa of the small intestine and encyst. Within the cyst they continue to grow for from 6 to 8 days, and then re-enter the lumen of the small intestine and develop into adults within a

few days. Hence, the complete cycle, from egg to egg, is approximately 15 days.

Several infected mice were kept as the source of eggs, and pans were used to collect the fecal pellets. Moist paper toweling covered the bottom of the pans, and wire-mesh screen was placed one inch above the toweling. The mice were placed in the pans and remained for a period of from 4 to 5 hours. The pellets were removed, comminuted in cool tap water, and strained through two thicknesses of cheese cloth. The mass was washed with tap water until 300 ml. of the filtrate were obtained. The filtrate was centrifuged, the supernatant discarded, and the sediment pooled, resuspended, and washed two more times. After the last washing the supernatant was discarded and enough water added to the sediment to make a suspension resembling the consistency of mineral oil.

Four pieces of Whatman No. 40 filter paper, 7.0 cms. in diameter, were placed in a petri dish. The sediment containing the eggs was streaked across the paper and enough water added to saturate the dry areas. The cultures were kept at a temperature of 21–25°C. The lids of the petri dishes were removed and the cultures aerated for 30 minutes each day (enough water was added to replace that lost to evaporation).

At the end of five days, the filter paper was held at an angle and the infective larvae washed off with a fine stream of water. They were collected with a pipette and placed in 50-ml. centrifuge tubes.

The procedures for standardization and infection were similar to those used for *Trichinella spiralis* as described by Larsh and Kent (1949) with one exception. In the present study, a cold (10°C.) aqueous solution of 5% white Karo syrup was used as the suspending medium.

Since the adult worms may persist for as long as eleven months after infection, it was necessary to remove them by drug treatment prior to a succeeding infection(s). Otherwise, it would not be possible, for certain, to separate the worms from the initial and later infection(s), and such counts could not, therefore, be used to measure immunity. Accordingly, each animal received 0.5 ml. of a 5% suspension of carbon tetrachloride (hereafter referred to as CCl_4) prepared in a solution of 0.2% plain agar in water (Hewitt, 1958). The drug was administered orally, using a blunt, slightly curved (18-gauge, 1 ¾-inch) needle.

The recovery and counting of adult worms followed, with certain modifications, the method of McGee *et al.* (1957). In this case, the intestine was not everted on an applicator stick, but was allowed to remain in the 1% HCl solution (56°C.) for 20 minutes. At this time, those worms not disengaged from the tissue were removed, under magnification (1.5X) by dissecting needles and tweezers. The intestine, now free of worms, was discarded and cold tap water (10°C.) was added to the petri dish to relax the worms. Adult counts were then made with the aid of a dissecting microscope fitted with 15X oculars and 0.66X objectives. The dish was placed on a sheet of ruled paper to facilitate counting and the total number of worms was determined by a direct count.

The criterion used to measure the degree of immunity was the number of adult worms present in the small intestine 15 days after the challenging infection.

The data from the experiments were analyzed statistically by Student's "t" and the analysis of variance tests. The calculations and tables of these determinations are on file at the Department of Parasitology, University of North Carolina.

Experimental Procedures and Results

Series One

EXPERIMENT I. It was the purpose of this experiment to determine whether one infection would stimulate a demonstrable immune response against *N. dubius*.

Sixteen mice were divided into two main groups with eight mice in each group. The mice of Group I (experimentals) received (*per os*) one stimulating infection with 50 *N. dubius* larvae. Twenty-one days later, they received a single oral dose of CCl_4 to remove the adult worms from the intestine. Four days after the drug treatment, the Group I mice and the non-stimulated controls (Group II) were given a challenging infection with 50 *N. dubius* larvae. Appropriate controls were used. These included: (1) viability controls (given the stimulating infection and sacrificed 15 days after infection to check on the infectivity of the larvae), (2) drug-action controls (given the stimulating infection, treated with CCl_4 and sacrificed 4 days later to check on the efficacy of the treatment), and (3) drug-residue controls (treated with CCl_4 and given the challenging infection 4 days later to determine whether or not the drug had a residual effect on this infection). These three sets of special controls were included in the remaining experiments of this series, but to conserve space they will not be described again.

The mice of Groups I and II (and the drug-residue controls) were necropsied 15 days after

Table I

The number of *N. dubius* recovered after a challenging infection with 50 larvae.

EXPERIMENT I		EXPERIMENT II		EXPERIMENT III	
Group I (Experimentals; received one stimulating infection)	Group II (Nonstimulated controls; received only the challenging infection)	Group I (Experimentals; received two stimulating infections)	Group II (Nonstimulated controls; received only the challenging infection)	Group I (Experimentals; received three stimulating infections)	Group II (Nonstimulated controls; received only the challenging infection)
33	33	40	43	4	42
40	50	22	41	8	29
37	47	28	29	7	48
43	38	32	40	12	31
36	33	27	38	17	40
32	37	24	31	18	34
47	36	41	41	19	35
38	38	26	30	17	37
Mean 38.2	39.0	30.0	36.6	12.7	37.0

the challenging infection and the number of adult *N. dubius* harbored in the small intestine was counted.

EXPERIMENTS II AND III. The next two experiments of this series were similar to the one above in purpose and design, except that different numbers of stimulating infections were used. The animals of Experiment II received two stimulating infections while those of Experiment III were given three stimulating infections at 21-day intervals.

RESULTS. In all three experiments, the number of worms recovered from the three sets of special controls showed that the various required conditions of the experiments were met. The viability controls (total of 24 in the three experiments) harbored the number of worms expected from each 50-dose infection, proving the infectivity of the larvae used in the various stimulating infections. The drug-action controls (8 in each experiment) harbored no worms, showing that the drug treatment was entirely successful in eliminating the worms of the various stimulating infections. Finally, the drug-residue controls harbored about the same number of worms as the nonstimulated controls (Group II), indicating that the drug given 4 days prior to the various challenging infections did not affect the number of worms recovered 15 days after these infections.

The average number of *N. dubius* adults recovered from the experimentals (Group I) and nonstimulated controls (Group II) for each experiment is shown in Table I.

Since the difference in the means of the two groups in Experiment I is 0.8, it is obvious there is no significant difference between the two groups. The findings indicate, therefore, that under these experimental conditions one stimulating infection did not produce a detectable degree of immunity.

In the second experiment, a definite, if not statistically significant, trend was noted in that fewer worms were recovered from the mice given two stimulating infections before being challenged. In this case, the experimentals (Group I) had an average of 30.0 worms, while the nonstimulated controls (Group II) had an average of 36.6 worms.

In the third experiment, a statistically significant (1% level) reduction in the number of adult worms was noted. In this case, the experimentals (Group I) had an average of 12.7 worms, while the nonstimulated controls (Group II) had an average of 37.0 worms. It is clear, therefore, that the three stimulating infections produced a striking immune response.

When the experimental animals (Group I) of Experiment III were necropsied 15 days after the challenging infection, it was evident that not all the worms were in the lumen of the small intestine. Gross examination revealed that a number of worms could be seen encysted within the intestinal tissue. By comparison, no worms were seen encysted in the tissues of the nonstimulated controls (Group II). This suggested that the immune mechanism(s) in some way might have delayed the entry of the worms into the lumen of the intestine. If so, a later necropsy period might reveal more adults than observed at 15 days. On the possibility that a later period might be more accurate in making comparisons between groups in future experi-

ments, the final experiment of this series was designed.

EXPERIMENT IV. This experiment was designed to determine the number of worms present in the intestinal lumen of immunized animals at 15, 30, and 60 days after a challenging infection.

Forty-eight mice were divided into 6 groups of 8 mice each. The experimentals (Groups I, II, and III) and the nonstimulated controls (Groups IV, V, and VI) were handled as above (Experiment III), except that experimental Groups II and III and the nonstimulated controls of Groups V and VI were sacrificed 30 and 60 days after the challenging infection, respectively.

RESULTS. The results for the special controls were similar to those given above for the first three experiments.

The average number of *N. dubius* adults recovered from the experimentals (Group I) and nonstimulated controls (Group II) is shown in Table II.

As shown in Table II, the experimental mice (Groups I, II, and III) had an average of 8.4, 13.3, and 10.8 worms, respectively, while the nonstimulated controls (Groups IV, V, and VI) had an average of 36.4, 39.4, and 33.7 worms, respectively.

An analysis of variance carried out with the data of this experiment showed a highly significant (1% level) variation between the worm recoveries from the different groups of mice. Hence, the variations between groups was due to the immunity developed in response to the three stimulating infections. On the other hand, comparisons between the various experimental groups did not indicate a significant difference, showing that the number of worms present at 15, 30, and 60 days after the challenging infection was similar. The same was shown by comparison between the three control groups. Therefore, since there was no significant change in the number of adult worms in the lumen of the small intestine between 15, 30, and 60 days, there seemed to be no justification for changing the necropsy date (15 days after the challenging infection).

Series Two

The degree of tissue contact during the life cycle of *N. dubius* is considerable. It seemed likely, therefore, as in the case of certain other parasites having both a parenteral and intestinal lumen phase, that most of the immunity demonstrated above is stimulated by the larvae during their tissue migration. In any event, it seemed worthwhile to consider tests to determine whether the adults and/or larvae are responsible for the immunity demonstrated.

Table II

The number of adult *N. dubius* recovered from 47 mice at necropsy 15, 30, and 60 days after a challenging infection with 50 larvae—Experiment IV. (Experimentals received 3 stimulating infections prior to the challenging infection; non-stimulated controls received only the challenging infection.)

EXPERIMENTALS			NONSTIMULATED CONTROLS		
Group I; 15 days	Group II; 30 days	Group III; 60 days	Group IV; 15 days	Group V; 30 days	Group VI; 60 days
1	3	0	42	39	41
9	4	2	39	50	33
6	9	19	25	36	27
11	2	26	32	40	49
9	33	3	30	30	21
4	20	34	45	45	28
12	27	2	30	36	40
15	8	1	48	*	31
Mean 8.4	13.3	10.8	36.4	39.4	33.7

* Died from undetermined cause(s).

For the present study, the life cycle was divided arbitrarily into two parts, identified as Phase I and II. Phase I of the life cycle has reference to the larvae (or pre-adults) used for infection, and begins, therefore, when the larvae penetrate from the lumen into the mucosa of the small intestine and includes the entire period of tissue habitation (about 9 days). Phase II refers to the adult worms, from the time they emerge from the tissue into the lumen until they are passed naturally (or after drug treatment) from the body. Hence, this includes the period of sexual maturity of the adults, copulation, and ovipositing activities of the females.

Drug treatment was employed to eliminate the adult worms as soon as possible after their appearance in the lumen of the intestine. This was necessary to eliminate one phase (Phase II) so that the relative importance of the other (in this case, Phase I) could be determined. However, before the method of drug treatment could be used, it was necessary to know precisely the time of emergence of the adults from the tissue into the intestinal lumen. Against the possibility that these periods vary as to the immune state of the host, it was necessary to make separate determinations after one, two, and three infections.

Table III

The average number of adult *N. dubius* recovered from the lumen of the small intestine of four mice after one, two, and three infections with 50 larvae.

Day no.	EXPERIMENT I (Experimentals; given one infection)	EXPERIMENT II (Experimentals; given two infections; worms of first infection removed with CCl_4)	EXPERIMENT III (Experimentals; given three infections; worms of first two infections removed with CCl_4)
1	0	0	0
2	0	0	0
3	0	0	0
4	0	0	0
5	0	0	0
6	0	0	0
7	1.2	3.0	0
8	25.5	13.7	0
9	33.2	25.0	0
10	42.5	32.5	3.7
11	45.0	34.5	18.2
12	39.0	36.0	12.2
13	43.2	35.2	23.5
14	40.7	43.5	25.2
15	43.7	35.7	12.8
16	38.5	39.2	12.2
17	41.2	36.0	23.0
18	40.7	38.0	12.5
19	42.0	39.7	31.5
20	41.2	39.0	24.2
21	40.2	37.0	35.7

EXPERIMENT I. This experiment was designed to determine the time of appearance of adult worms (Phase II) in the intestinal lumen after one infection with 50 larvae.

Eighty-eight mice were used. The 84 experimentals and the 4 persistency controls were given one infection. Twenty-four hours after infection, and at 24-hour intervals for 21 consecutive days, 4 experimental animals were sacrificed and adult counts were made. On day 21, the persistency controls were also sacrificed.

EXPERIMENTS II AND III. The next two experiments were similar to the one above in purpose and design, except that different numbers of infections were used. The animals of Experiment II received two infections, while those of Experiment III received three infections. It was, of course, necessary to remove (with CCl_4) each infection before giving a succeeding one, since these worms may persist for months. Drug-action controls were used to check on the effectiveness of the various treatments. In Experiment II, drug treatment was given 21 days after the first infection and 4 drug-action controls were sacrificed 4 days later. The second infection was given 25 days after the first infection. In Experiment III, drug treatment was administered 21 days after both the first and second infections and the 4 drug-action controls in each case were sacrificed 4 days after treatment. By these procedures, it was possible to determine the time of appearance of the adult worms in the intestinal lumen after two (Experiment II) and three (Experiment III) infections.

RESULTS. The number of worms recovered from the persistency controls was similar to that from the experimentals on day 21, showing that the worms persisted in usual numbers. No more than 4 worms were recovered from any of the three sets of drug-action controls, proving that the drug was effective in removing most of the worms of each prior infection.

The average number of adult worms recovered from the groups of experimental mice for each experiment is shown in Table III. As seen in Table III, few worms were noted in the first two experiments before 8 days. Furthermore, it is noted that there was little difference in the average number of worms recovered between 11 and 21 days. However, in the third experiment few worms appeared before 11 days, and

there was not a great difference in the average number of worms recovered between 14 and 21 days.

From these results, it is clear that almost all of the worms emerged from the tissue into the intestinal lumen between 8 and 21 days after the various infections.

To eliminate the worms before there would be much opportunity to stimulate an immune response(s), it appeared that CCl_4 would have to be given 8 days after one or two infections and 11 days after a third infection. To eliminate worms' emerging after these periods, it seemed that a second treatment would have to be given 3 days after the first in the hope that almost all of the worms emerging after 8 days would be eliminated. To test this method of drug treatment, the next three experiments of this series were designed.

EXPERIMENT IV. This experiment was designed to determine whether or not two treatments with CCl_4, administered 8 and 11 days after one infection, will eliminate the adult worms (Phase II).

Thirty mice were divided into two main groups of 15 mice each. The Group I mice (experimentals) and the Group II mice (controls) were given an infection with 50 larvae. The Group I mice were treated with CCl_4 8 and again at 11 days after infection. On day 9 (24 hours after first treatment), on day 12 (24 hours after second treatment), and on day 21 (10 days after second treatment), 5 mice from each group (I and II) were sacrificed and adult counts were made of the number of adults recovered from the small intestine.

EXPERIMENTS V AND VI. These were performed concurrently with Experiment IV, and differed from it only in the number of infections given. In Experiment V, two infections with 50 larvae each were given at 21-day intervals. The CCl_4 treatments were given at 8 and 11 days after both infections. It was not necessary to include viability controls and drug-action controls for the first infection, since the results of Experiment IV provided this information. Five mice each were sacrificed on days 9, 12, and 21 after the second infection. In Experiment VI, three infections were given with 50 larvae each at 21-day intervals. The CCl_4 treatments were given at 8 and 11 days after the first two infections and at 11 and 14 days after the third infection. Again, viability controls and drug-action controls were not needed, since the results for Experiments IV and V provided this information. Five mice each were sacrificed on days 12, 15, and 21 after the third infection.

RESULTS. The number of adult *N. dubius* recovered from the small intestine is shown in Table IV.

From the results of these three experiments, it is clear that the two treatments with CCl_4 removed almost all of the adult worms from one, two, and three infections. Moreover, in view of the emergence studies (Table III), the periods selected should have prevented no more that a few worms from remaining in the small intestine for any period of time. Thus, it seemed that this method would be satisfactory for use in determining whether or not the adult worms in the lumen of the small intestine play a demonstrable role in the development of acquired immunity. Admittedly, the method does not meet entirely the conditions of this design, since a few worms were able to persist for several days. However, it required three stimulating infections of 50 larvae each to produce a demonstrable immunity (Table I), and, in this case, all of the adults were permitted to remain in the lumen for 21 days after each stimulating infection. Thus, for practical purposes, it would seem permissible to disregard the few worms persisting after use of this method, as it is highly unlikely that their presence could influence the immunity to a demonstrable degree.

EXPERIMENT VII. This experiment was designed to show whether or not the adult worms (Phase II) in the lumen of the small intestine play a demonstrable role in the production of immunity to *N. dubius*.

Twenty-four mice were divided equally into 3 main groups. The 8 Group I mice (experimentals) were given three stimulating infections of 50 *N. dubius* larvae each at 21-day intervals, while the Group II mice (nonstimulated controls) were not infected. Eight and 11 days after the first and again after the second infection, and 11 and 14 days after the third infection, the experimentals were treated with CCl_4. Seven days after the last treatment (or 21 days after the third infection), the mice of both groups were given a challenging infection with 50 larvae. Necropsy was performed 15 days after challenge, and adult worm counts were made. The remaining 8 mice were placed in Group III (drug-residue controls). They were needed to determine whether or not the 6 drug treatments given after the stimulating infections had a residual effect on the worms developing from the challenging infection. Hence, they received the same drug treatments

Table IV

The number of *N. dubius* adults recovered from the small intestine of treated and untreated mice at various intervals after one (Experiment IV), two (Experiment V), and three (Experiment VI) infections (50 larvae each).

EXPERIMENT IV		EXPERIMENT V		EXPERIMENT VI	
Group I (Experimentals; one infection; CCl_4 8 and 11 days after infection)	Group II (Controls; one infection; no treatment)	Group I (Experimentals; two infections; CCl_4 8 and 11 days after each infection)	Group II (Controls; two infections; worms of first infection removed with CCl_4 21 days after infection)	Group I (Experimentals; three infections; CCl_4 8 and 11 days after first two infections, and 11 and 14 days after third infection)	Group II (Controls; three infections; worms of first two infections removed with CCl_4 21 days after each infection)
Day 10	36	Day 10	14	Day 0	5
9 11	28	9 12	20	12 0	25
5	33	14	16	1	20
7	43	5	17	0	10
6	35	7	19	2	8
Mean 7.8	35.0	9.6	17.2	0.6	13.6
Day 0	48	Day 9	34	Day 3	20
12 0	38	12 0	26	15 2	32
0	33	5	41	3	22
0	43	0	34	1	21
0	35	1	37	4	24
Mean 0	39.4	3.0	34.4	2.6	23.8
Day 0	42	Day 3	*	Day 2	25
21 0	45	21 0	34	21 4	30
2	46	2	31	1	25
0	39	3	40	5	34
0	38	0	32	3	23
Mean 0.4	42.0	1.6	34.2	3.0	27.4

* Died from undetermined cause(s).

as the Group I mice, but were given only the challenging infection. Appropriate viability and drug-action controls were included as described above.

RESULTS. The numbers of worms recovered from the various viability controls and from the various drug-action controls were those expected, proving that the larvae used for the various stimulating infections were of the usual infectivity and that the drug removed almost all of the adults from these various infections.

The average number of *N. dubius* adults recovered from the groups of experimental and control mice is shown in Table V. The experimentals (Group I) had an average of 11.2 worms while the nonstimulated controls had an average of 42.5 worms. The drug-residue controls (Group III) had an average of 40.4 worms.

An analysis of variance with the data of this experiment showed a highly significant (1% level) difference between the groups of mice. The analysis further indicated that the difference was due to the immunity developed in response to the three stimulating infections. On the other hand, the differences between the two control groups (II and III) were not significant.

It is understood that it was not possible, by the methods used, to eliminate 100% of the adult worms as they emerged from the tissue into the lumen of the small intestine. Thus, the adults (Phase II) could have contributed to the immunity demonstrated. However, considering the small numbers involved and the

brief period of persistency it seems highly unlikely that this phase could have contributed enough to influence the results. It seems safe to conclude, therefore, that the adults (Phase II) did not play a demonstrable role in the immunity demonstrated in this way. This agrees with other studies on the lumen phase of parasitic worms and suggests strongly that most of the immunity demonstrated was due to the larval stages in the tissues.

Discussion

The experiments of Series One showed: (1) that a demonstrable immunity was produced in mice infected with *Nematospiroides dubius*, and (2) that there was no significant difference in the number of adult worms recovered from immunized mice between 15 and 60 days after a challenging infection.

In the first three experiments, acquired immunity was demonstrated only in mice that had received three stimulating infections prior to the challenging infection. The failure to demonstrate immunity in those receiving one or two stimulating infections suggests that the immunity has a quantitative basis as demonstrated in other host-parasite combinations (Larsh, 1946; Hendricks, 1950; Hurley, 1959).

It seems clear that the immune mechanism(s) was not aroused sufficiently by one or two stimulating infections to cause a significant reduction in the number of adults recovered after the challenging infection. There was no significant reduction of worms in mice given only one prior infection, although an apparent, if not significant, trend in reduced numbers of worms was noted in those given two stimulating infections. Finally, after three prior infections the reduction in numbers of worms was striking (Table I). Several workers have reported failure in attempts to produce an active immunity against *N. dubius* in mice (Spurlock, 1943; Baker, 1955; Liu and Ivey, 1958), but it is worth noting that, in all cases, the mice were given only one stimulating infection.

The final experiment of this series showed no significant difference in the number of worms recovered between 15 and 60 days after a challenging infection given to immunized mice (Table II). Thus, the worms observed in the intestinal tissue at 15 days did not succeed in reaching the intestinal lumen within the period of observation. It was observed that the worms were surrounded by a cyst wall as noted by Baker (1954). In the present case, however, these encysted forms were not seen except in mice that had been immunized with three stimulating infections. Hence, the results suggest that the worms were trapped by the defensive mechanism(s). Perhaps the reaction is similar to that described by Taliaferro and Sarles (1939) for *Nippostrongylus muris* in immunized rats.

The experiments of Series Two revealed: (1) the time periods after one, two, and three separate infections when the adults (Phase II) appeared in the lumen of the small intestine; (2) that almost all of the adults (Phase II) from one, two, and three separate infections were removed within a brief period of time by giving two CCl_4 treatments; and (3) that there was a striking immunity developed after three stimulating infections despite prompt removal of the adults by CCl_4 treatment after each of the three infections.

In the first three experiments, the time of appearance of the adults (Phase II) in the lumen of the small intestine varied between 7 and 21 days after infection. After one and after two infections, very few worms appeared before 8 days, and there was not a great difference in the average number of worms recovered between 11 and 21 days. After three infections, very few worms appeared before 11 days and there was not a great deal of difference in the average number of worms recovered between 14 and 21 days (Table III). The fact that in the latter case the first appearance was 3 days later than in the previous cases suggests that the worms were delayed in the tissues due to the action(s) of the immune mechanism(s).

The next three experiments determined that two treatments with CCl_4 would remove promptly almost all of the adult worms (Phase II) from the intestinal lumen. After one and two infections, treatment on days 8 and 11 removed almost all of the adults within a brief period. However, after three infections, it was necessary to delay the first treatment for 3 days as explained above. In this case, treatments given on days 11 and 14 were very effective in removing the adult worms (Table IV).

In the final experiment, a striking degree of immunity was produced in mice given three stimulating infections and treated with CCl_4 to remove almost all of the adults (Phase II) soon after their emergence into the intestinal lumen (Table V). This immunity was similar to that produced when the adults from all three stimulating infections were permitted to persist for 21 days (Table I). Therefore, these results offer strong evidence that the lumen-dwelling adults (Phase II) did not play a demonstrable role in this immunity. The same has been noted in studies with certain other parasites (Miller, 1932; Luttermoser,

Table V

The number of adult *N. dubius* recovered 15 days after a challenging infection with 50 larvae.

Group I (Experimentals; given three stimulating infections with 50 larvae each, given CCl_4 on days 8 and 11 of the first two infections, and on days 11 and 14 of the third infection)	Group II (Nonstimulated controls; given challenging infection only; no treatment)	Group III (Drug residue controls; given CCl_4 at same periods as mice of Group I; given challenging infection only)
10	47	40
4	38	38
18	48	42
11	40	39
5	37	43
2	35	32
33	53	45
7	42	44
Mean 11.2	42.5	40.4

1938; Hearin, 1941). Thus, it seems safe to suggest that the immunity demonstrated in this case was produced in response to the tissue-invading larvae (Phase I). Since this stage lasts approximately 9 days, one might expect immunity to be manifested after one infection. However, the immunity probably has a quantitative basis. If so, the size of the stimulating dose used here (50 larvae) may have been too small to produce changes sufficient to repel the same number of worms (50) in the challenging infection.

Summary

White mice given one stimulating infection with 50 *N. dubius* larvae failed to develop acquired immunity, as measured by adult worm counts 15 days after a challenging infection with 50 larvae. After two stimulating infections (50 larvae each), given at 21-day intervals, the mice also failed to show an acquired immunity by this criterion, but there was a trend toward smaller numbers of worms than in the non-stimulated controls. However, after three stimulating infections at the same intervals (21 days), the mice demonstrated a striking immunity. These results suggest a quantitative basis for the immunity, which probably accounts for earlier reported failures to produce a demonstrable immunity after one stimulating infection.

In mice immunized with three stimulating infections, a number of worms were noted encysted in the intestinal tissue at 15 days after the challenging infection. When the necropsy dates were extended to 30 and 60 days, there was no significant increase in the number of worms reaching the intestinal lumen; hence it appears that the encysted worms are trapped by the defensive mechanisms(s).

A striking degree of immunity was demonstrated in mice given three stimulating infections and treated with CCl_4 to remove almost all of the adult worms (Phase II) soon after their emergence into the intestinal lumen. These results suggest that the adults did not play a demonstrable role in the immunity demonstrated, providing strong evidence that the tissue-invading larvae (Phase I) accounted for the major part of the immunity.

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LITERATURE CITED

AFRICA, C. M. 1931. Studies on the host relations of *Nippostrongylus muris*, with special reference to age resistance and acquired immunity. Jour. Parasitol. **18**: 1–13.

BAKER, N. F. 1954. Trichostrongylidosis—the mouse as an experimental animal. Proc. Am. Vet. Med. Assoc. (Ninety-first ann. meeting, Seattle, Wash., Aug. 23–26), 185–192.

———. 1955. The pathogenesis of trichostrongyloid parasites: some effects of *Nematospiroides dubius* on the erythrocyte pattern and spleens of mice. Exper. Parasitol. **4**: 526–541.

EHRENFORD, F. A. 1954. The life cycle of *Nematospiroides dubius* Baylis (Nematoda: Heligmosomidae). Jour. Parasitol. **40**: 480–481.

HEARIN, J. T. 1941. Studies on the acquired immunity to the dwarf tapeworm, *Hymenolepis nana* var. *fraterna*, in the mouse host. Am. Jour. Hyg. **33** (Sect. D.): 71–87.

HENDRICKS, J. R. 1950. The relationship between precipitin titer and number of *Trichinella spiralis* in the intestinal tract of mice following test infections. Jour. Immunol. **64**: 173–177.

HEWITT, R. 1958. Personal Communication. Research Division, Experimental Therapeutics Research Section, Lederle Laboratories Division, American Cyanamid Company, Pearl River, New York.

HURLEY, F. J. 1959. Immunization threshold in laboratory rats given a small initial infection of

Nippostrongylus muris. Helminthol. Soc. Wash. **25**: 91–96.

KERR, K. B. 1938. Attempts to induce an artificial immunity against the dog hookworm, *Ancylostoma caninum*, and the pig ascaris, *Ascaris lumbricoides suum*. Am. Jour. Hyg. **27**: 52–59.

LARSH, J. E., JR. 1946. The effect of alcohol on development of acquired immunity to *Hymenolepis* in mice. Jour. Parasitol. **32**: 72–78.

LARSH, J. E., JR., AND D. E. KENT. 1949. The effect of alcohol on natural and acquired immunity of mice to infection with *Trichinella spiralis*. Jour. Parasitol. **34**: 45–53.

LIU, G. Y. H. AND M. H. IVEY. 1958. Preliminary studies on *Nematospiroides dubius* (Baylis, 1926). Jour. Parasitol. **44** (Suppl.): 32.

LUTTERMOSER, G. W. 1938. Susceptibility of chickens to reinfection with *Raillietina cesticellus* as determined by the presence of the original terminal segment. Jour. Parasitol. **24** (Suppl.): 14.

MCCOY, O. R. 1931. Immunity of rats to reinfection with *Trichinella spiralis*. Am. Jour. Hyg. **21**: 484–494.

MCGEE, G., R. RUSSOMANNO, AND J. H. SANDGROUND. 1957. An expeditious method for counting trichostrongylids and other helminths of the small intestine. Jour. Parasitol. **43**: 315–317.

MILLER, H. M., JR. 1932. Superinfection of cats with *Taenia taeniaformis*. Jour. Prev. Med. **6**: 17–29.

OTTO, G. F., AND K. B. KERR. 1939. The immunization of dogs against hookworm, *Ancylostoma caninum*, by subcutaneous injection of graded doses of living larvae. Am. Jour. Hyg. **29** (Sect. D.): 25–45.

SARLES, M. P. 1932. Development of an acquired resistance in rabbits by repeated infection with an intestinal nematode, *Trichostrongylus calcaratus* Ransom, 1911. Jour. Parasitol. **18**: 61–82.

SCHWARTZ, B., J. E. ALICATA, AND J. T. LUCKER. 1931. Resistance of rats to superinfections with a nematode, *Nippostrongylus muris*, and an apparently similar resistance of horses to superinfection with nematodes. Jour. Wash. Acad. Sci. **21**: 259–261.

SHELDON, A. J. 1937. Studies on active acquired resistance, natural and artificial, in the rat to infection with *Strongyloides ratti*. Am. Jour. Hyg. **25**: 53–65.

SPURLOCK, G. M. 1943. Observations on host parasite relations with laboratory mice and *Nematospiroides dubius*. Jour. Parasitol. **29**: 303–311.

STOLL, N. R. 1929. Studies with the strongyloid nematode: *Haemonchus contortus* I. Acquired resistance of hosts under natural reinfection conditions out-of-doors. Am. Jour. Hyg. **10**: 384–418.

TALIAFERRO, W. H. AND M. P. SARLES. 1939. The cellular reactions in the skin, lungs, and intestine of normal and immune rats after infection with *Nippostrongylus muris*. Jour. Infect. Dis. **64**: 157–192.

TAXONOMIC NOTES ON FISHES OF THE INTERIOR OF THE UNGAVA PENINSULA

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The interesting collection forming the basis of the present report was secured by Francis Harper during a trip in Labrador and Quebec in 1953. The trip was supported by the Arctic Institute of North America (under contractual arrangements with the Office of Naval Research) and by the Research and Development Division, Office of the Surgeon General, Department of the Army. The specimens, numbering 130, were collected from June to September.

The geographical positions of the collecting localities are shown in the introduction to the paper by Dr. Harper, which immediately follows this one. The length given for the various specimens is the total length, unless otherwise specified.

Salmonidae

Salmo salar ouananiche McCarthy
Figure 1 (No. 662).

The following specimens were taken in the Ashuanipi River, one to two miles north of Ashuanipi Lake. No. 662 (315 mm.), August 24. No. 666 (a head only, 155.5 mm. long), August 29. No. 674 (116 mm.), September 6; few black spots on back. No. 679 (275 mm.), August or September; fewer black spots on back and sides than in No. 662.

Cristivomer namaycush (Walbaum)

No. 605 (436 mm.), outlet of Knob Lake, June 28; the pale spots, though variable in size, are all more or less distinctly rounded. No. 619 (210 mm.), Attikamagen Lake, Iron

Arm, July 8; pale spots and markings large and variable. No. 633 (215 mm.), river at north end of Lake Wapanikskan, July 25.

Salvelinus fontinalis (Mitchill)

Nos. 602, 603, and 604 (232, 185, and 170 mm.), Sucker Creek, June 27; in No. 602, pale spots on back numerous and very closely set; in No. 604, much fewer than in the larger specimens. Nos. 612, 616, 618, 625, 626, and 627 (164, 34, 147, 40, 38, and 36 mm.), Attikamagen Lake, Iron Arm, July 4 to 12; some were taken in the shallow shore waters, others in an inlet brook; pale spots few in No. 618. No. 632 (255 mm.), river at north end of Lake Wapanikskan, July 25. Nos. 645, 646, and 647 (63, 55, and 64 mm.), Camp Brook, August 6. No. 653 (185 mm.), outlet of Mollie T. Lake, August 9; with seven obscure blackish parr marks. No. 658 (283 mm.), outlet of Leroy Lake, August 20; pale spots far more numerous than in smaller specimens; about eight or ten, at least, above lateral line. No. 675 (148 mm.), Ashuanipi River, September 6; parr marks distinct.

Coregonidae

Coregonus clupeaformis (Mitchill)

Nos. 598, 599, and 600 (443, 250, and 204 mm.), Sucker Creek, June 25. No. 634 (401 mm.), Lac Aulneau, July 26. Nos. 656 and 657 (371 and 310 mm.), Mollie T. Lake, August 13 and 14. Nos. 669, 670, and 671 (412, 300, and 255 mm.), Ashuanipi Lake, August 30. Nos. 672 and 677 (330 and 210 mm.), Ashuanipi River, one mile north of Ashuanipi Lake, September 2 and 10.

Scale counts of eight specimens from the Hamilton Basin (Sucker Creek, Ashuanipi Lake, and Ashuanipi River) average 77.8 in lateral line to caudal base and 2.6 more on latter; 10.9 above, 9.5 below to ventral origin, 9.4 to anal origin. Scale counts of three specimens from the Koksoak Basin (Lac Aulneau and Mollie T. Lake) average 77.3 in lateral line to caudal base and 1.7 more on latter; 10.3 above, 9 below to ventral origin, 8.3 to anal origin.

Specimens in the former group have less deep bodies (depth 4.1 to 4.6 in standard length) than those in the latter group (depth 3.6 to 4.0) (*cf.* Plate 1, Fig. 2, and Plate 1, Fig. 5, in the following paper). Thus the former approach *C. labradoricus* (Richardson) in having a body tending toward the shape of *Prosopium*. Richardson (1836: 206) remarks on this character in describing *labradoricus* from the "Musquaw" (=Musquaro) River, a tributary of the Gulf of St. Lawrence. The length of his type specimen (14 inches) was fairly comparable with that of several of the above-mentioned specimens from Ashuanipi Lake and River.

Prosopium cylindraceum quadrilaterale (Richardson)

No. 601 (290 mm.), Sucker Creek, June 27; scales 86 in lateral line to caudal base and 3 more on latter; 10 above, 9 below to ventral origin; 8 below to anal origin. No. 678 (178 mm.), Ashuanipi River, September 9; scales 83–4; 10–8–7.

Esocidae

Esox lucius Linnaeus

No. 648 (a head only, 185 mm. long), Guy's River, August 6; differs from a specimen[1] from Nueltin Lake, Keewatin, in that the lower

[1] Proc. Acad. Nat. Sci. Philadelphia **100**: 152, fig. 21, 1948.

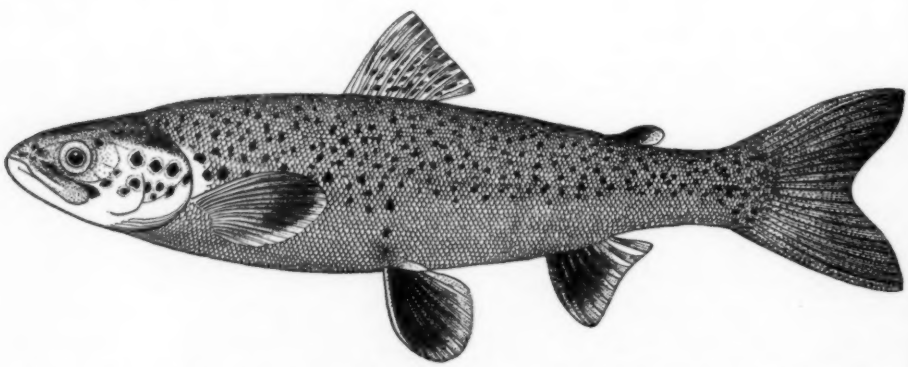

FIG. 1. *Salmo salar ouananiche*. No. 622, 315 mm. Ashuanipi River, two miles north of Ashuanipi Lake, Labrador. August 24, 1953. H. W. F. del.

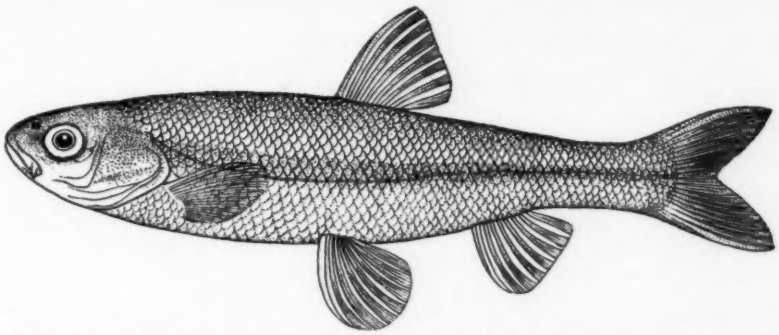

FIG. 2. *Couesius plumbeus.* No. 660, 120 mm. Ashuanipi Lake, Labrador. August 24, 1953. H. W.

sides of the head have more buff or pale brown lines; the large eye is 4 in the snout.

Catostomidae

Catostomus catostomus catostomus (Forster)

No. 593 (four specimens, largest 38 mm.), Knob Lake, June 15. Nos. 594 and 606 (35 and 38 mm.), Knob Lake, June 21 and 28. No. 659 (49 specimens, largest 29 mm.), Ashuanipi Lake, August 24; uniform pale olive, little paler below; no dark markings; fins pale.

Nos. 665 and 673 (363 and 382 mm.), Ashuanipi River, August 28 and September 2. In the first the papillae on the lower lip are far larger than shown in my figure[2] of a specimen from Nueltin Lake, Keewatin. The Labrador specimen shows 8 in a transverse (if irregular) row. Moreover, the pectoral fin of this specimen is shorter and does not reach more than halfway to the ventral. The fish is without pearl organs and does not show their scars or impressions. The second specimen is much like the first.

Cyprinidae

Couesius plumbeus (Agassiz)
Figure 2 (No. 660)

No. 595 (three specimens, 28-33 mm.), Knob Lake, June 21. Nos. 611 and 617 (24 and 27 mm.), Attikamagen Lake, Iron Arm, July 2 and 7. Nos. 640–644 (83–110 mm.), Camp Brook, August 6. Nos. 660 and 661 (120 and 116 mm.), Ashuanipi Lake, August 24.

In all the specimens their dull colors suggest changes due apparently to the preservative (formalin). The blackish lateral stripe is distinct on the snout, extends under the eye, and is most prominent on the caudal peduncle.

Gadidae

Lota lota lacustris (Walbaum)

Nos. 596, 597, 649, 650, and 651 (78, 81, 101, 93, and 118 mm.), Knob Lake, June 22 and 23, July 4 and 6. No. 680 (16 mm.), Attikamagen Lake, Iron Arm, July 12.

Gasterosteidae

Gasterosteus aculeatus Linnaeus

Nos. 609, 610, 620, and 621 (46-63 mm.), Attikamagen Lake, Iron Arm, July 2 and 12. Nos. 628, 629, and 630 (31–65 mm.), Attikamagen Lake, Northwest Bay, July 17. Nos. 637, 638, and 639 (34–53 mm.), Lac Aulneau, July 27 and August 2.

Cottidae

Cottus cognatus Richardson

No. 652 (67 mm.), Knob Lake, July 4. Nos. 615, 622, 623, and 624 (32–58 mm.), Attikamagen Lake, Iron Arm, July 7 and 12. No. 631 (31 mm.), Attikamagen Lake, Northwest Bay, July 18. No. 676 (65 mm.), Carol Lake, September 17.

[2] Proc. Acad. Nat. Sci. Philadelphia **100**: 143, fig. 2, 1948.

FIELD AND HISTORICAL NOTES ON FRESH-WATER FISHES OF THE UNGAVA PENINSULA AND ON CERTAIN MARINE FISHES OF THE NORTH SHORE OF THE GULF OF ST. LAWRENCE[1]

FRANCIS HARPER

Introduction

PREVIOUS INVESTIGATIONS. Among the few nineteenth-century writers on the fishes of the Ungava Peninsula, scarcely one published anything approaching a full account of the species. The majority confined their attention to those of the coastal waters, and more especially to those of economic importance, such as the cod, salmon, capelin, lake and brook trouts, and arctic char. In this category of incomplete recorders, endowed with something less than a taxonomic ichthyologist's viewpoint, we may place Cormack (1826), Fortin (1862–1865), Hind (1863), Hamilton (1865), Weiz (1866), Chimmo (1868), Bell (1879; 1886), and Holme (1888). To this period also belongs McLean (1849; 1932), who was the discoverer of Grand Falls and the first white man to traverse the northeastern part of the peninsula; he devoted three lines to four fishes of Ungava.

Another group, still restricting their attention mainly to coastal waters, but with the definite status of zoologists, included Sir John Richardson (1836), Storer (1850), Stearns (1883), and Packard (1866; 1891). Richardson described *Coregonus labradoricus* from the "Musquaw" (=Musquaro) River; Storer described several new species from Nova Scotia and Labrador; Stearns provided a briefly annotated list (mainly from the North Shore of the Gulf of St. Lawrence); and Packard did likewise, extending his area through the Strait of Belle Isle and northward up the Atlantic Coast.

The most voluminous and the most valuable nineteenth-century account of the fishes of the Ungava Peninsula is doubtless one prepared by Lucien M. Turner (1885) as a result of his sojourn at Fort Chimo from 1882 to 1884. Unfortunately it has remained merely in manuscript form, in the library of the United States National Museum, but there is prospect of its eventual publication (see Dunbar and Hildebrand, 1952: 85). Turner's notes include such topics as native names of fishes and methods of capture, food habits, distribution, frequency, spawning season, and color.

In the last decade of the century two writers, Chambers (1896) and Low (1896), drew aside the veil of secrecy that for so long a time had obscured the species of the interior waters. In his monographic work on the ouananiche, the former brought out a considerable fund of information on other fresh-water fishes (mainly game species) of the Lake St. John area, on the southwestern border of the peninsula. Low, among the results of his remarkable exploratory journeys in various directions across the peninsula in the 1880's and 1890's, gave us (1896) the first comprehensive, though brief, published account of the fish life of the far interior; his interests, however, were focused more particularly on the food and game species, with consequent neglect of such small fry as minnows, sticklebacks, and sculpins. For half a century or so afterward, Low remained our primary authority on the fishes of the interior wilds.

Only a little professional collecting is evident during the first two decades of the present century. Evermann and Kendall (1902) summed up our knowledge of the distribution of species in the St. Lawrence River, but their territory lay for the most part beyond the limits of the Ungava Peninsula. Evermann and Goldsborough (1907) provided a useful check list of the fresh-water fishes of all Canada, including such distributional information as was available at that time. Kendall (1909; 1910) reported on two collections from the east coast, the first made by the Bowdoin College Expedition of 1891 and the second by Owen Bryant.

After their exploration of the Naskaupi and George River basins, Mrs. Hubbard (1908) and Wallace (1906; 1907) furnished incidental

[1] Biological investigations in this region in 1953 were supported by the Arctic Institute of North America (under contractual arrangements with the Office of Naval Research) and by the Research and Development Division, Office of The Surgeon General, Department of the Army. The results are being prepared for publication under a grant from the National Science Foundation.

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EDITOR'S NOTE. Dr. Harper has declined to accept certain editorial suggestions. While the Journal does not agree with his style and usage at many points, it has acceded to his wishes as far as possible.

Table I

	Latitude	Longitude	Altitude (ft.)
Labrador			
Abel Lake	54°46′ N.	66°47′ W.	1,624
Ashuanipi Lake, N. end	53°00′ N.	66°15′ W.	1,750
Ashuanipi River, 1–2 mi. N. of Ashuanipi Lake	53°02′ N.	66°15′ W.	1,740
Attikamagen Lake, Iron Arm, NW. end	54°56′ N.	66°39′ W.	1,536
Attikamagen Lake, Northwest Bay	54°59′ N.	66°41′ W.	1,536
Carol Lake	53°04′ N.	66°58′ W.	2,000
Guy's River	54°43′ N.	66°19′ W.	1,520
Howell's River (mouth)	54°35′ N.	66°40′ W.	1,510
Shabogamo Lake	53°15′ N.	66°30′ W.	1,700
Snelgrove Lake	54°37′ N.	65°49′ W.	1,567
Stevens Lake	53°05′ N.	66°57′ W.	2,200
Sucker Creek (tributary to Abel Lake)	54°46′ N.	66°48′ W.	1,630
Quebec			
Al's Lake	55°17′ N.	67°13′ W.	1,500
Camp Brook (tributary to Knob Lake)	54°48′ N.	66°50′ W.	1,650
Chubb Crater	61°17′ N.	73°40′ W.	2,200
Eric Lake	51°55′ N.	65°40′ W.	1,925
Fort McKenzie	56°50′ N.	68°57′ W.	220
Hanna's Lake	54°49′ N.	66°42′ W.	1,540
Knob Lake	54°48′ N.	66°49′ W.	1,645
Lac Aulneau	57°01′ N.	68°38′ W.	510
Lac Matemace	54°54′ N.	66°49′ W.	1,535
Lac de Morhiban	51°50′ N.	62°53′ W.	1,720
Lac Ochiltrie	52°35′ N.	73°15′ W.	1,000?
Lake Wapanikskan	56°57′ N.	68°55′ W.	720
Larch River (mouth)	57°40′ N.	69°30′ W.	50
Leroy Lake	55°08′ N.	67°14′ W.	1,590
Mollie T. Lake	55°02′ N.	67°09′ W.	1,625
Nemiscau River (headwaters)	51°48′ N.	75°00′ W.	1,100
Peter's Lake	54°32′ N.	66°45′ W.	1,535
Pearce Lake	54°49′ N.	66°49′ W.	1,630
Rivière du Gué (mouth)	57°21′ N.	70°45′ W.	500?
Scott Lake	55°11′ N.	67°24′ W.	1,775
Seven Islands	50°12′ N.	66°23′ W.	0
Thunder River	50°17′ N.	64°45′ W.	0

notes on some of the food and game species. Cabot (1912) did the same for the coastal region about Davis Inlet and for some of the neighboring fresh waters. Comeau, an outstanding resident naturalist of the North Shore of the Gulf, supplied (1909; 1923) particularly interesting information on the life history of the salmon. Flaherty (1918) provided a few ichthyological notes pertaining to his two traverses between Hudson and Ungava bays.

The period since 1930 has witnessed a resurgence of activity in the labors of professional ichthyologists directed toward the Ungava Peninsula, although approximately half of those about to be mentioned have not themselves penetrated the interior. Dymond has published on the coregonine fishes of Hudson and James bays (1933) and on the Atlantic salmon in Ungava Bay (1941), and in addition he has brought out a check list of the fresh-water fishes of Canada east of the Rocky Mountains (1947). Vladykov has published on the fishes of Hudson Bay (1933) and on lampreys (1949). Weed (1934) has written on the sea trouts. The salmon in Labrador is discussed by Blair (1943*a* and 1943*b*), and the capelin by Templeman (1948). L. R. Richardson (1944) gives an account of the species of "central northern" (more prop-

erly, southwestern) Quebec. Legendre and Rousseau (1949) list 10 species of the George River and Payne River areas. Polunin (1949) gives a few incidental notes on trout and other species (not always identifiable) in northern areas. Munroe (1949) presents a useful though incomplete account of the species of the Knob Lake area. Backus (1951) publishes some new and rare records from coastal waters, and in a more comprehensive work (1957) he provides an excellent report on his extensive researches in Labrador (both coastal and inland), together with a review of earlier records. Dunbar and Hildebrand (1952) present a most informative account of the species of the Ungava Bay area. Wynne-Edwards (1952) includes various fresh-water species of the peninsula in his review of those of the Arctic and Subarctic in general. Legendre (1954) brings out an illustrated key to the fresh-water fishes of Quebec. Martin (1955) contributes notes on those of the Chubb Crater area. Oliver (1958) reports on the fishes of Astray Lake and other nearby waters, and McAllister and Bleakney (1959) publish on fresh-water species of northeastern Quebec and Labrador.

This brief review makes no pretension of completeness, especially as to marine investigations.

LOCALITIES. Since some of the interior localities from which material or information were obtained in 1953 are not named on existing maps, the list in Table I is provided, showing the latitude, the longitude, and at least the approximate altitude of all the localities for hitherto unpublished records. This list is not expanded to cover additional localities referred to in citing records from the works of others. Many of the dots on the distributional maps represent locality records derived from the previous literature but not necessarily stated in detail in the present report. Nearly all such localities, however, may be found in the works cited in connection with the summation of previous records of each species.

I have ventured to apply the unofficial names Sucker Creek and Camp Brook to two small streams where several observations were made. They are northwesterly tributaries of Abel Lake and Knob Lake, respectively.

This paper deals primarily with the fishes of the interior of the peninsula, where my personal observations and collections extended from Ashuanipi Lake in the south to Lac Aulneau in the north. I have, however, included notes on several maritime species (*Petromyzon, Salmo salar salar, Mallotus, Gadus*, and *Cyclopterus*), concerning which information was obtained in the vicinity of Seven Islands, on the North Shore of the Gulf.

DISTRIBUTION OF FISHES OF THE UNGAVA PENINSULA BY DRAINAGE BASINS. The vast Ungava Peninsula covers an extent of territory approximately equal to a quadrilateral in the eastern United States whose four corners are Chicago, New York, Jacksonville, and New Orleans. The southwestern limits of the peninsula may be placed at the Saguenay and Nottaway Rivers, as indicated by a broken line on Map 1. It is divisible into four main drainage basins or watersheds—southern, eastern, northern, and western; or, in other words, the St. Lawrence drainage, the Atlantic drainage, the Hudson Strait and Ungava Bay drainage, and the Hudson and James bays drainage. Some of the principal streams of the first are the Saguenay, Betsiamites, Outardes, Manicouagan, Marguerite, Moisie, Magpie, Romaine, Natashquan, Little Mecatina, St. Augustin, and St. Paul rivers; of the Atlantic drainage, the Paradise, Eagle, Hamilton, Naskaupi, Canairitok, Assiwaban (or Koraluk), and Fraser rivers; of the Hudson Strait and Ungava Bay drainage, the George, Whale, Koksoak, Leaf, and Payne rivers; of the Hudson and James bays drainage, the Povungnituk, Kogaluk, Nastapoka, Little Whale, Great Whale, Fort George, East Main, Rupert, and Nottaway rivers. As far as distributional and biological aspects of the fish life are concerned, the western watershed is probably the least known of the four, although it appears to possess the greatest number of species.

It may be interesting to compare the fresh-water fish faunas of these four drainage basins and to note the comparatively few differences in their composition. Of the thirty-four species and subspecies listed in Table II, eight are recorded from all four basins; seven from three basins; eight from two basins; and ten from only one basin. Various taxonomic uncertainties and gaps in distributional information hinder the presentation of a clearer picture. The number of species and subspecies fairly definitely known from each basin varies only from 17 to 22. As field investigations are continued, a greater uniformity than is now apparent in the distribution of species in the various watersheds may be shown. It is quite likely that several of the river systems have interlocking headwaters, enabling fishes to pass from one drainage basin to another. Low (1896: 188, 159) indicates that such a connection exists at Summit Lake, a common source of the Kaniapiskau and Manicouagan rivers;

Table II

Distribution of fresh-water and anadromous fishes of the Ungava Peninsula by drainage basins.

	St. Lawrence	Atlantic	Hudson Strait and Ungava Bay	Hudson and James Bays
Petromyzon marinus	+	−	−	−
Acipenser oxyrhynchus	−	+	−	−
Acipenser fulvescens	−	−	−	+
Salmo salar salar	+	+	+	−
Salmo salar ouananiche	+	+	+	−
Cristivomer namaycush	+	+	+	+
Salvelinus fontinalis	+	+	+	+
Salvelinus alpinus	−	+	+	+
Leucichthys artedi	−	−	+	+
Coregonus atikameg atikameg	−	−	−	+
Coregonus clupeaformis	+	+	+	+
Prosopium cylindraceum quadrilaterale	+	+	+	−
Esox lucius	+	+	+	+
Catostomus commersonii commersonii	+	+	+	+
Catostomus catostomus catostomus	+	+	+	+
Catostomus catostomus nanomyzon	+	−	−	−
Moxostoma aureolum	−	−	−	+
Semotilus corporalis	+	−	−	+
Semotilus atromaculatus atromaculatus	−	−	−	+
Couesius plumbeus plumbeus	+	+	+	+
Rhinichthys atratulus atratulus	−	−	−	+
Rhinichthys cataractae	+	+	−	+
Anguilla rostrata	+	+	−	−
Lota lota lacustris	+	+	+	+
Eucalia inconstans	−	−	−	+
Pungitius pungitius	−	−	+	+
Gasterosteus aculeatus	+	+	+	−
Percopsis omiscomaycus	−	−	−	+
Perca flavescens	+	−	−	+
Stizostedion vitreum vitreum	+	−	−	+
Cottus bairdii bairdii	+	+	−	−
Cottus bairdii kumlieni	−	+	−	−
Cottus cognatus (cognatus)	−	+	+	−
Cottus cognatus gracilis	−	+	+	−
Total	20	20	17	22

+Recorded. −Not recorded.

and that Michikamau Lake, in times of high water, discharges into the Hamilton River as well as into the Naskaupi. Moreover, it would appear from two maps in the National Topographic Series that Myrtle Lake may be another body of water lying athwart a major divide, discharging at one end into the Koksoak system and at the other into the Hamilton system. It is situated close to the Northwest Bay of Attikamagen Lake (Knob Lake sheet, east half, and Tait Lake sheet, east half).

It is worthy of note that both *Salmo salar salar* and *S. s. ouananiche* are lacking from only the western watershed, and *Salvelinus alpinus* from only the southern watershed. *Coregonus a. atikameg* is known, within the peninsular limits, from only the shore waters and lower river courses of the Hudson and James bays drainage, and *Moxostoma, Rhinichthys a. atratulus, Semotilus a. atromaculatus, Percopsis,* and *Eucalia* from only the upper waters of that drainage. *Semotilus corporalis, Perca,* and *Stizostedion* seem to occur only in the southern and western watersheds. Whether these distributional similarities and differences are the result of geological history, of varying

temperature requirements, or of still other factors, are questions beyond the general scope of the present paper.

Perhaps, however, a bit of speculation on the ouananiche is permissible. This fish, as a derivative of *Salmo salar salar*, has apparently been unable to populate the rivers and lakes draining into the east side of Hudson and James bays, owing to the absence of the salmon itself from those bays. It would also appear that there are no interlocking headwaters on the divide separating this drainage basin from the St. Lawrence and the Hudson Strait–Ungava Bay basins, since the ouananiche does not seem to have surmounted that divide.

The known localities from which the various species have been recorded are indicated by the larger dots on the accompanying distributional maps (nos. 1–20), which have been compiled from the published literature and from new information offered in the present paper. The boundaries between the Canadian, Hudsonian, and Arctic zones are approximately indicated by bands of tiny dots. The amelioration of the climate in recent decades seems to have pushed the boundary between the Canadian and the Hudsonian zones to a considerable distance north of the line indicated by Merriam *et al.* (1910) (see Harper, 1958: 19–20, 26, Map 1, and 1961: 15–19, Map 2).

A number of the fishes listed in Table II do not appear in the systematic accounts of species on subsequent pages, being unknown as yet in that part of the interior covered by my investigations. They are the following:

Acipenser oxyrhynchus	Atlantic Sturgeon
Acipenser fulvescens	Lake Sturgeon
Salvelinus alpinus	Arctic Char
Moxostoma aureolum	Northern Redhorse
Rhinichthys atratulus atratulus	Black-nosed Dace
Rhinichthys cataractae	Long-nosed Dace
Semotilus corporalis	Fallfish
Semotilus atromaculatus atromaculatus	Northern Creek Chub
Percopsis omiscomaycus	Trout-perch
Perca flavescens	Yellow Perch
Eucalia inconstans	Brook Stickleback
Pungitius pungitius	Nine-spined Stickleback

The distributional status of these fishes may be ascertained from such works as Low (1896), Vladykov (1933), L. R. Richardson (1944), Scott (1954), Backus (1957), or Slastenenko (1958).

ECONOMIC UTILIZATION. Hitherto there appears to have been practically no utilization of fishes from the interior wilds of Ungava as a food resource for the outside world. Up to the beginning of mining developments a few years age, virtually the only ones who took advantage of the abundance of food fishes were scattered Indian families and an occasional exploring party of geologists or naturalists. Along with the recent influx of mining and construction personnel, many enthusiastic fishermen among them have cast their flies and trolls in the lakes and rivers and have been abundantly rewarded, without causing appreciable depletion among the species they sought (principally lake trout, brook trout, and ouananiche). The Indian families have no doubt depended mainly upon gill-nets for securing a winter's supply of fish. In view of the present inadequacy of the big-game resources of the central part of the peninsula, fishes must play an important part in the food economy of the native trappers.

I neither saw nor heard of any organized commercial fishing in the interior in 1953. This happy state of affairs is likely to be modified in the future. The completion of the railway from Seven Islands to Knob Lake in 1954 made the lakes adjacent to the railway line available for commercial fishing, with shipment of products by rail and steamer to various cities of Canada and the United States. Hopefully, these operations will be subject to such regulations as will prevent the serious depletion of any waters and thus safeguard them as a permanent source of excellent food.

MEASUREMENTS. In some cases my own measurement of the length of a given specimen, made at the time of capture, is slightly larger than Fowler's (1961) measurement of the same specimen, made several years later. The shrinkage is doubtless due to the action of the preserving fluid.

ACKNOWLEDGMENTS. The generous support of the Arctic Institute of North America, the Office of Naval Research, the Office of the Surgeon General of the Army, and the National Science Foundation, which made my Ungava investigations possible, is most gratefully acknowledged. These investigations were very greatly facilitated by the fine hospitality of the Iron Ore Company of Canada. From headquarters at Knob Lake I traveled in various directions to spend from one to several weeks at a time with different field parties of this company and with one party of Holannah Mines, Ltd. For information or assistance of various kinds in the field (including the presentation of specimens), I am deeply indebted to the following persons: A. E. Moss, Richard Geren, J. M. Harrison, R. D. Macdonald, D.

O. Macdonald, F. D. Foster, Cyrille Dufresne, Peter Almond, Greg Anderson, Ron Barrett, William Bazuk, Herb Chrysler, Pierre Côté, Antoine Deschênes, James Espinard, Fred Farah, Roger Ferguson, W. Foisy, Robert Girardin, Guy Giroux, Charles Grace, Myron Kazmirchuk, Joseph Laporte, Edward Lawson, Philippe Loth, Kai Mansa, Peter Midlige, John Macko, James Murdoch, H. E. Neal, R. Gordon Racey, William Sehrøpfer, Gilbert Simard, Robert Slipp, Norman Sliter, James Thompson, J. L. Véronneau, Lance Widnall, and Ross Wilson.

I must thank a number of Montagnais friends, not only for an interesting list of fish names in their own tongue (which are included in the following account of species), but also for specimens and general information. They are Sebastien, Francis, and Ben McKenzie; Jérôme St. Onge; Willé, Kom, Bastien, and Michel Pinette; Georges Michel; and Raymond Beaudin. Père J. E. Beaudet, O. M. I., has most kindly checked the Montagnais names for me. Very few of these names are found in Lemoine (1901), and then with a somewhat different spelling (see under *Esox* and *Anguilla*).

M. J. Dunbar, of McGill University, and E. H. Grainger, of the Fisheries Research Board of Canada, furthered my investigations in various ways. Edward C. Raney, of Cornell University, has very kindly read the manuscript, giving me the benefit of many useful suggestions and criticisms. C. B. Wurtz has been good enough to identify mollusks in the stomach contents of certain fishes.

For collecting permits I am greatly indebted to the Province of Quebec Department of Game and Fisheries and to the Province of Newfoundland Department of Mines and Resources. I brought back 130 specimens representing eleven fresh-water species, and also the stomachs of twenty-seven additional specimens. This material has been, or will be, divided between the Fisheries Research Board of Canada, the United States National Museum, Cornell University, and the Academy of Natural Sciences of Philadelphia.

The basic outline of Maps 1–20 was redrawn, with permission of the Geological Survey of Canada, from Map 1045A, Geological Map of Canada (1955).

Accounts of Species

• *Petromyzon marinus* Linnaeus. Sea lamprey; lamproie de mer (Fr.)

Charles Grace spoke of having noted large numbers of "lamprey eels" in the Moisie River at Mile 13 of the Q. N. S. & L. Railway; this was probably in June or early July of 1951. He referred to their buccal funnel and gill openings. He estimated their length at 18 to 24 inches. The species was presumably *Petromyzon*.

It was possibly the same species that Low (1896: 329) reported as "a small Lamprey (*Petromyzon* sp.)" found adhering to a brook trout in the Bersimis River in 1884. Bigelow and Welsh (1925: 18) include Labrador in the range of *Petromyzon*. Vladykov (1949: 45) states that it occurs in the entire Gulf of St. Lawrence and ascends the St. Lawrence River and its main tributaries. These are perhaps the principal records from the Ungava Peninsula.

• *Salmo salar salar* Linnaeus. Atlantic salmon; saumon atlantique (Fr.); otchatchomek (M.). Map 1.

Several miles east of Seven Islands, on May 30, I noticed a salmon net extending from the shore out into the Gulf of St. Lawrence. It was made of strong twine, with a mesh of about 7½ inches. The fisherman tending it spoke of having gotten two salmon from it.

Sebastien McKenzie obtained salmon while he stayed one year at the junction of Larch River and the Rivière de Gué. He said they ascended Larch River to about longitude 72°. To that point there are apparently no falls that would obstruct the upstream migration of the salmon. This report is in accord with Low (1898: 15) concerning the presence of salmon in that part of the Larch (or Stillwater) River and in the Gué (or Kenogamistuk) River.

The range of the salmon includes the entire south coast of the peninsula (Cayley, 1863: 91; Low, 1886: 7; Lucas, 1891: 722; Chambers, 1896: 136–138; Evermann and Kendall, 1902: 237; Bryant, 1913: 9; Townsend, 1913: 173; Cabot, 1920: 319; Comeau, 1923: 19, 33, 51, 175; Préfontaine and Cuerrier, 1944: 106; Vladykov, 1946: 26, 60); the east coast north at least to Ramah (Holme, 1888: 190, 193, 200; Packard, 1891: 399; Low, 1896: 329; Cabot, 1912: 52, 58, 93, 115, 195, 213; Grenfell, 1922: 328; Blair, 1943a and 1943b; Tanner, 1947, 1: 433; Bigelow and Schroeder, 1953: 121; Legendre, 1954: map following p. 12; Backus, 1957: 287); and the north coast—George, Whale, Koksoak, and Leaf rivers (Low, 1896: 122, 329; Dunbar and Hildebrand, 1952: 92); lower Larch and Kaniapiskau rivers (Power, 1958: 63). Dunbar and Hildebrand place the normal western limit on Ungava Bay at the

Koksoak River. Within the range thus outlined, the salmon undertakes spawning migrations from the sea up the rivers until it encounters unsurmountable falls. The time of these migrations extends from June on the St. Lawrence coast to August on Ungava Bay (Low, 1896: 330); thus they seem to be at least roughly synchronized with the onshore migrations of the capelin. According to Bouthillier (1948: 24), in the St. Lawrence Basin the salmon no longer ascends any rivers west of the Isle aux Coudres (about 50 miles below Quebec City). It is unknown in Hudson Bay.

The Clearwater River, from which Oliver (1956) records the species, is not the one flowing into Richmond Gulf (as he informs me, *in litt.*, November 30, 1959), but a tributary of the Larch River.

Cartwright recorded 12,396 salmon taken at White Bear River (on the east coast) between June 23 and July 20, with an average weight of 15 pounds (Hind, 1863, 2: 193). Hind also (1863, 2: 239) mentions a maximum length of 3 feet 11 inches and a maximum weight of 40¼ pounds in the Moisie River. Comeau (1923: 380–416) supplies an interesting record of the total numbers and weights of salmon taken on the Godbout River during a fifty-year period (1859–1908), together with certain additional records from the St. John, Moisie, and Trout rivers (pp. 368–379).

• *Salmo salar ouananiche* McCarthy. Ouananiche (Eng., Fr., and M.). Map 2.

"Ouananiche" is evidently a Montagnais word taken over as both the French and the English name of the species. The explanation that its meaning is "little salmon" (the terminal *iche* signifying diminutive) encounters a certain difficulty from the fact that the Montagnais word for salmon is *ouchachoumac* at Lake St. John (Chambers, 1896: 40) and *otchatchomek* at Seven Islands. For further discussion of the derivation, see McCarthy (1894: 206), Chambers (1896: 39–57), and Montpetit (1897: 498).

On June 13 A. E. Moss brought from Guy's River a ouananiche weighing 3½ pounds. The only other specimens I saw during the season were taken between August 23 and September 6 in the vicinity of some rapids on the Ashuanipi River about 2 miles north of Ashuanipi Lake. This area (Plate 2, Fig. 4) was a favorite resort of fishermen from the big construction camp at Mile 224 Airstrip; and it furnished the devotees of rod and reel with a plentiful supply. Lance Widnall, who presented me with several specimens, stated that the species spawns in the rivers at places where there is a good current (but not in the rapids), where the water is from two to four feet deep, and where there are good-sized stones on the bottom. These conditions exist in the Ashuanipi River; and the water is clear. On September 6 Mr. Widnall reported several large males and females that had not yet spawned. (In the Lake St. John area this activity takes place in October, according to Chambers [1896: 13].) Seven individuals from about 11 to 16 inches in length, which I examined on August 27 and 29, were not in spawning condition; their stomachs contained fish and insects (E. H. Grainger). A male caught on August 29 (Plate 1, Fig. 4), about 25½ inches in length and weighing 7¾ pounds, was the largest specimen that I observed; it exhibited the elongate and hooked mandible characteristic of salmon in general at the spawning season. Its lunate or slightly emarginate tail is in marked contrast to the forked tails of the two smaller specimens in the same figure. Another male of almost equal size, taken on August 23, had much-enlarged testes. In each of these adult males the stomach was empty. The largest individual recorded by Chambers (1896: 104, 195) was an eight-pounder from Lake Tschotagama, northeast of Lake St. John. Thus the maximum size of the ouananiche falls far short of that of the Atlantic salmon.

The following color notes were made on a fresh 118-mm. specimen taken and presented by James Thompson on September 6: dusky dorsally, silvery laterally, whitish ventrally; about 9 parr marks from 5 to 7 mm. broad, moderately distinct though faint when taken out of water; sides (mostly above the lateral line) with the regular adult dusky spotting; in addition, about 11 or 12 red spots, fairly evenly spaced along the lateral line; head dusky above, brassy-silvery on opercle, white below; small dusky spots on chin and throat; dorsal and caudal fins faintly tinged with yellowish, the former with two rows of dusky spots; adipose with dusky punctulations and a narrow orange-red border; anal with dusky punctulations along rays, general color yellowish, becoming orange-red toward apex; ventrals plain, tinged with yellowish, becoming pale orange-red toward apex; pectorals yellowish with numerous dusky punctulations; mandible dusky on anterior half.

Richard Geren reported the ouananiche as common in the fall in Howell's River, a northwesterly affluent of Astray Lake; and Philippe Loth reported it in Lac de Morhiban.

Low (1896: 182, 330) records the species in Hamilton River above Grand Falls; in Kaniapiskau River below Lake Kaniapiskau; in the Romaine, Manicouagan, and upper George rivers; in Michikamau and Mushalagan lakes; and in Lake St. John. Additional localities mentioned by Chambers (1896: 12, 13, 19, 112, 114, 222) are various rivers and lakes tributary to Lake St. John; the Ashuanipi river; the Goynish, the Musquaro, and probably many other rivers that flow into the lower Gulf of St. Lawrence. Some ouananiche in Lake Tschotagama had their stomachs full of sticklebacks (Chambers, 1896: 195). Wallace (1907: 51, 52) found the species in Crooked River, which discharges into Grand Lake. Cabot (1920: 310) reports it in Minipi Lake; Blair (1943*a*: 4, 11), from the headwaters of the Naskaupi River downward and in Wabush Lake; Backus (1957: 287), in Flour Lake; Oliver (1958: 103), in Astray Lake. Munroe (1949: 169) speaks of it as apparently rare in the Menihek lakes area, and mentions a specimen from Guy's River. Just as the salmon seems not to enter Hudson Bay, so its derivative the ouananiche is unknown in the lakes and streams tributary to that body of water (Low, 1896: 182). Legendre's distributional map (1954: following p. 12) indicates only a single locality north and east of Lake St. John!

• *Cristivomer namaycush* (Walbaum). Lake trout; touladi, toulazi, or touradi (Fr.); kokemesh or kukumess (M.); kokomish (Naskapi, *fide* Weed, 1934). Map 3.

If Chambers (1896: 270) is correct in considering "touladi" a Micmac and Abenaki name for the lake trout, it would appear to be only secondarily a French name, taken over from the native speech.

This fine fish is of very general distribution in the fresh waters of the Ungava Peninsula, not only north to the limit of trees but even beyond, as in Payne Lake and in the Chubb Crater area. With the possible exception of the pike, it is the largest game fish of the far interior, attaining several times the weight of the ouananiche. It is also more widely and more thoroughly distributed than the ouananiche. As far as my limited observations went, the ouananiche exceeded the lake trout in numbers only in the Ashuanipi River. While the main habitat of the present species is evidently the lakes, it occurs in many rivers as well. The waters in which it is found are clear, generally with rocky bottoms; most of the rivers have a rapid current.

The French name *touladi* (or its variants) seems, as a rule, to be applied rather indiscriminately to all lake trout. On the other hand, at Ashuanipi Lake, Ross MacFarland expressed the opinion that a fish so named is not just an ordinary lake trout, but one with a large head and a slender, undersized body. At times he has hesitated or even declined to eat such a fish.

Most of the specimens that I saw from June 6 to September 16 were taken by trolling; a few others, in gill-nets. They came from the following waters: Aulneau (Plate 1, Fig. 5), Leroy, Mollie T., Attikamagen, Knob, and Carol lakes (Plate 1, Fig. 1); a river at the north end of Lake Wapanikskan; outlet of Knob Lake; Guy's River; and Ashuanipi River below Ashuanipi Lake (Plate 1, Fig. 4). The species was reported by J. M. Harrison in a lake near Chubb Crater; by J. L. Véronneau (*in litt.*, August 29, 1953, and January 26, 1956) in Scott Lake and in Lac Ochiltrie or vicinity; by H. E. Neal in Shabogamo Lake; by Willé Pinette in Eric Lake; and by Philippe Loth in Lac de Morhiban. These are the statistics for some of the larger specimens that I examined: 10 pounds, 32 inches in length, Mollie T. Lake; 24 pounds and 14½ pounds, Guy's River or vicinity; and 18½ pounds, about 33 inches in length, Ashuanipi River. There was a report of a 37-inch specimen, weighing 18 pounds, from the Ashuanipi River.

Stomach contents were noted as follows: June 7, lake south of Knob Lake, an unidentified fish about 4 inches long from a 19-inch lake trout; June 10, Knob Lake, five small trout (sp.?) from a 21-inch individual; June 10, vicinity of Knob Lake, remains of a small fish from an 11½-pound individual; July 25, river at north end of Lake Wapanikskan, about six mice (apparently meadow mice, *Microtus*) from a 23-inch individual; about August 4, Mollie T. Lake, a round whitefish about 8 inches long, from a large individual; September 12, Carol Lake, a mouse (probably *Microtus*) from a 6½-lb. individual. Furthermore, H. E. Neal reported three "baby muskrats" (possibly adult *Microtus*?) found in a lake trout at Shabogamo Lake during the summer.

In possible explanation of the numbers of mice consumed, it may be remarked that both meadow mice and red-backed mice (*Clethrionomys*) had reached an unusually high population peak in the interior of the country in 1953. Three *Microtus* (two fresh, one decomposed) were found floating in Carol Lake in September; evidently the mice both at Lake

Wapanikskan and at Carol Lake had felt some urge to take to the water and had there succumbed, either from natural causes or as prey to the lake trout. H. Hildebrand (1949: 311) reports shrews in the stomachs of lake trout in the George River area, and Dunbar and Hildebrand (1952: 95) report mice as well.

An 11½-pound female, taken in the Knob Lake area on June 10, had fairly developed ovaries. A specimen at Lac Aulneau, July 26 (Plate 1, Fig. 5), contained eggs about 2 mm. in diameter. Another, which was taken in Leroy Lake on August 18 and weighed 8½ pounds, was full of roe. A male, taken in the Ashuanipi River on August 29 and weighing 5⅛ pounds, had exceeding small testes. On September 6 Lance Widnall said that the spawning season should have been over by that time.

A fresh 1¼-pound specimen, taken in the outlet of Knob Lake on June 28, furnished the following color notes: dusky olive on dorsum with paler vermiculations; sides dusky olive with roundish silvery or very pale grayish spots; underparts from chin to caudal fin white; dorsal, adipose, and caudal fins dusky with vermiculations; caudal very pale orange distally, dull orange along ventral edge; ventrals and anal orange-red, margined outwardly with whitish; pectorals dusky, pale and uncolored on outer margin, very pale orange on inner margin; head dusky, with irregular silvery spots on opercle; jaws marked with dusky; iris dusky orange. In a 21-inch specimen taken at Knob Lake on June 10, the spots on the sides were not golden. Among a bushel or so of lake trout (the largest weighing 14½ pounds) taken at Guy's River or vicinity on June 30, the spots on the sides were small and rather silvery, not golden. An 18½-pounder, taken in Ashuanipi River on September 3, was more golden-colored than any other lake trout I had seen in the Ungava region.

Some plant material and a few trematode eggs (identified by Aaron Goldberg) were found in the intestine of a 10-pound specimen taken in Mollie T. Lake on August 15. I did not detect gill parasites in various specimens examined.

Low (1886: 7) records the lake trout from Betsiamites River and Pipmuacan Lake and also (1896: 56, 70, 101, 137, 182, 330) from the larger lakes of the interior north to Hudson Strait. Among these lakes are Mistassini (which yielded a maximum weight of 40 pounds), Nichicun, Winokapau, Mushalagan, and Michikamau, and lakes of the upper Hamilton Basin. On a later trip Low (1898: 11, 12) found lake trout in Clearwater River and Lake. Chambers (1896: 267) specifies "nearly all the lakes of the Labrador peninsula," including Lake St. John, Lake Tschotagama, and Lac à Jim. Obalski (1905: 20) records the species from Chibougiche, Obatogamau, and Chibougamau lakes. Evermann and Goldsborough (1907: 102) add Manuan Lake and Mingan River, and Kendall (1909: 210) reports the Northwest River. Wallace (1906: 121, 149) mentions Lost Trail Lake and Windbound Lake and again (1907: 80, 108) Namayeush Lake and Lake Kasheshebogamog along the route from Northwest River to Michikamau Lake. Prichard (1910: 692) found the species in most of the lakes between the Fraser River and Indian House Lake. Cabot (1912: 101, 228, 269) gives records from the Assiwaban River and the outlet of Mistinipi Lake; it is the principal fish, he says, of the Naskapi Indians of the George River area. Flaherty (1918: 120) reports it from Lake Minto; Weed (1934: 127), between Hopedale and Nain; and Blair (1943*a*: 11), from Wabush Lake. L. R. Richardson (1944: 207) has this fish feeding on *Cottus* and *Leucichthys* in Lake Wakonichi. Legendre and Rousseau (1949: 134–135) record it from George and Kogaluk rivers and Payne Lake; Polunin (1949: 48, 169, 173, 182, Plate 16), from Lac Bienville and McGill Lake and vicinity; and Pomerleau (1950: 13), from the headwaters of the Manicouagan River. Munroe (1949: 169) specifies Wishart Lake and Guy's River in the Hamilton drainage, and Boundary, Sunspot, and Hanna's lakes in the Koksoak drainage; he places spawning in the first half of September. Dunbar and Hildebrand (1952: 94) mention occurrence in the Koksoak River, in several lakes near Fort Chimo, in Leaf River, and in the sea at Nain; they emphasize the importance of the species to the Naskapi Indians. Bateman (1953: 3) records it south of Leaf Bay. Bleakney (1954) writes of the food (largely mosquitoes and their larvae) in Squaw Lake. Martin (1955: 491) collected the species in the Chubb Crater area, and Oliver (1958: 103), in Astray Lake. *L'Action Catholique* (Quebec) of December 9, 1956, published a photograph of a number of *Cristivomer* taken on the Toulnustouc River. McAllister and Bleakney (1959: 33) report the species from Squaw Lake and Fort McKenzie.

• *Salvelinus fontinalis* (Mitchill). Brook trout, speckled trout; omble (truite) moucheté, truite saumonée (Fr.); matamek (M.). Map 4.

This species occurs quite generally throughout the Ungava Peninsula, north to Payne Lake. I obtained records from Aulneau (Plate 1, Fig. 5), Attikamagen, Pearce, Knob, and Stevens lakes and from the following streams: river at north end of Lake Wapanikskan, outlets of Leroy and Mollie T. Lakes, a small tributary of the Iron Arm of Attikamagen Lake, Guy's River or vicinity, Camp Brook, Sucker Creek, an inlet of Carol Lake, and Ashuanipi River. The species was also reported in Lac Matemace (Greg Anderson) and in Lac de Morhiban (Philippe Loth). Thus it occurs in waters varying from the larger lakes and rivers to brooks a few feet in width; the streams are characterized by clear water, rocky to muddy bottom, and swift to moderate current. Most of the specimens I saw were taken on a fly, but one was taken in a gill-net and juveniles in a dipnet. The general abundance of brook trout in Ungava waters, with their excellent flavor and the comparative ease of securing them with the fly, make them a frequent and very acceptable addition to the fare of field parties.

Some of the largest individuals examined (all from Guy's River or vicinity) had the following weights: 4½, 4, 2½, and 2¼ pounds. The smallest specimens seen (Sucker Creek, June 12) appeared about an inch (25 mm.) in length; four from Attikamagen Lake (July 7–12) were from 34 to 40 mm. and three from Camp Brook (August 6) were from 55 to 64 mm.; these figures give some indication of the rate of growth in northern waters during the brook trout's first year of life. At Knob Lake an individual of about 5 inches was seen on June 7, and others, about 9 or 10 inches long, on June 7 and 10; six taken in the outlet of Mollie T. Lake on August 10 had lengths of about 8 to 11 inches; three, of about 8 or 9 inches, were taken in Ashuanipi River on August 27; and one of about 7 inches was noted in Stevens Lake on September 11. It is quite probable that different rates of growth obtain in different waters.

A 255-mm. individual from Pearce Lake, June 28, had consumed insects. One adult and three subadults, taken at Attikamagen Lake on July 4, had eaten insects and fish, including a sculpin (*Cottus*). Six individuals, from 8 to 11 inches long, taken in the outlet of Mollie T. Lake on August 10, had swallowed insects and mollusks, including two *Lymnaea* sp.; in the stomach of a 10-inch individual was the gravel-encrusted case of a caddis worm. (The general abundance of caddis flies in Ungava should make their larvae a widely available food.) The stomachs of three individuals about 8 or 9 inches long, which had been taken in Ashuanipi River on August 27, contained insects and tapeworms. (The insect, fish, and tapeworm material reported here was identified chiefly by E. H. Grainger; the mollusks, by C. B. Wurtz.)

Three adults (from 170 to 232 mm.), taken in Sucker Creek on June 27, had venters faintly to strongly tinged with reddish. A freshly caught, 255-mm. male, taken in Pearce Lake on June 28, provided the following color description: dorsum dusky, mottled with pale olive; sides silvery bluish, with numerous roundish, whitish-gray spots up to 3 mm. in diameter; also with smaller and less numerous vermilion spots above and below lateral line, some within and some without the larger pale spots; under parts, chin to tail, silvery white; dorsal fin mottled and spotted with dusky, faintly yellowish posteriorly; adipose with a large central dusky spot; caudal obscurely marked with dusky and faintly tinged with very pale orange, the lower border faintly reddish; anal with a few dusky marks and pale orange-red anteriorly, the anterior margin white; ventrals plain posteriorly, orange-red anteriorly, separated from white anterior margin by a dusky streak; pectorals plain posteriorly, dusky anteriorly, the whole very faintly tinged with pale orange except for whitish anterior margin; head dusky above, its sides silvery with very faint orange tinge; margin of maxillary colorless; mandible dusky.

A 4½-pounder, taken in the vicinity of Guy's River on June 30, was orange-red on the lower sides of the body; white on under side of head, on breast, and on mid-venter; numerous vermilion and pale golden spots on sides; pectorals, ventrals, and anal rather bright red, partly dusky toward the white margin; caudal dull reddish.

A 36-mm. specimen, taken in Attikamagen Lake on July 7, exhibited about twelve transverse dusky parr marks on the sides, two of them broken in two; basal portion of caudal pale orange, divided by a broad blackish blotch; anal tinged with very pale orange, and ventrals and pectorals with a very faint suggestion of this color; pectorals and caudal with dusky punctulations.

Among approximately twenty-seven brook trout taken in a river close to Lake Wapanikskan on July 25 and averaging scarcely a pound

(maximum probably about 1½ pounds), some were blackish and reddish on the belly. Half a dozen large individuals (up to about 4 pounds), which were taken in the vicinity of Guy's River on August 6, were an even more brilliant red than those seen previously. Out of a bucketful taken from the outlet of Mollie T. Lake on August 9, a few were quite red-bellied, others not. The next day, the reddish-bellied individuals from the same waters were males, but some of the females approaching spawning condition were somewhat tinged with red on the under parts. The specimens taken in the outlet of Leroy Lake on August 20 were darker and more brilliant generally than individuals from other localities—the venter more blackish and reddish.

Several specimens from Ashuanipi River, seen in late August and on September 1, had pale bellies—not rich red and black as in more northern waters. On the other hand, two about 14 or 15 inches long, which were seen in an inlet of Carol Lake on September 12, were very black-bellied. According to Greg Anderson, the brook trout of Lac Matemace are distinguished from others of nearby waters by larger size, redder flesh, and more brilliant coloration (also, apparently, by numerous gill parasites). The foregoing records indicate that while the coloration becomes more intense at the breeding season, it varies more or less with the locality.

A female, taken on June 30 in the vicinity of Guy's River, had eggs 1 to 2 mm. in diameter. Fish of both sexes, taken from the outlet of Mollie T. Lake on August 10, exhibited enlarged gonads (one female had ova up to 5 mm. in diameter). On September 6 Lance Widnall considered that the brook trout of the Ashuanipi area were about to spawn.

On June 7, in Knob Lake just above the outlet, a school of approximately 35 *Couesius plumbeus*, which were 5 or 6 inches long, was accompanied by a slightly smaller brook trout; also two brook trout 9 or 10 inches long were accompanied by a round whitefish of about equal size. On June 12 one or more brook trout were associating with large schools of red suckers in Sucker Creek. On August 6 three brook trout (from 55 to 64 mm. long) and five plumbeous minnows (from 83 to 110 mm. long) were collected in the same short stretch of Camp Brook.

Out of five or six specimens from the outlet of Mollie T. Lake, August 10, every one had more or less numerous intestinal parasites (*Echinorhynchus leidyi* [Acanthocephala]).

The sea-run individuals of this species, found in the mouths of rivers and in adjacent coastal waters, have at times been distinguished by the name of *Salvelinus fontinalis hudsonicus* (Suckley). According to Jordan (1929: 52), this race "is silver-gray, and nearly plain, and reaches a large size; . . . spots orange rather than vermilion." In the following summation of records from the peninsula, it will not generally be distinguished from the fresh-water form unless by the indication of habitat.

Storer (1850: 264) indicates all streams of the southern Labrador coast as the brook trout's habitat; he records "water mice" from stomachs. Hind (1863, 1: 143–144, 209) found this species in tributaries of the Moisie River. Packard (1866: 275) mentions Square Island; and Couper (1868: 6, 11) reports "sea-trout" in Little Watshishu River and at the mouth of the Natashquan. Bell (1886: 12) reports the capture of brook trout by Eskimos south of Cape Wolstenholme. Low (1886: 7, 17) presents records from Betsiamites River and Lakes Pipmuacan and Mistassini; and later (1896: 56, 70, 137, 331), from many lakes (including Winokapau) and rivers (including Romaine, upper Hamilton, and Northwest); he also records sea-run fish from various localities, but in at least some cases confuses *Salvelinus fontinalis* with *S. alpinus* (see Backus, 1957: 292). Low (1898: 11, 12) also reports brook trout in Clearwater River and Lake. Chambers (1896: 97, 209, 232, 243, 246, 249) describes the fishing in Lake St. John and various neighboring waters: Little Peribonca and Ouiatchouan rivers, Lac de Belle Rivière, and Lake Batiscan. Wallace (1906: *passim*) refers to this trout in Susan River, Goose Creek, Beaver River, Lake Disappointment, Lake Mary, and Windbound Lake; and again (1907: 34, 39, 53, 69), near the lower Naskaupi River, in Crooked River, and near Lake Nipishish. Mrs. Hubbard (1908: 114) found it in Michikamau Lake; and Durgin (1908: 78), in the lower Eagle River. Kendall (1909: 210) adds Red and Chateau bays; Gathorne-Hardy (1911: 222, 224) mentions lakes near Hopedale and lower Fraser River; and Comeau (1923: 353), the Godbout River. Cabot (1912: 137, 153, 159, 228, 233, 251, and plate opposite p. 112) took this trout in the Assiwaban River area, finding mice in the stomachs in 1905, when these little animals had reached a population peak; he also (1920: 310, 319, 320, 329) reports it in the Coxipi, Eskimo (=St. Paul), Paradise, and St. Augustin rivers. Vladykov (1933: 6) reports it at Cape Jones, Long Point, and near Seal River, James Bay, and at Great Whale River, and to the north thereof; Weed (1934: 127–131) records it along the whole east coast north to Nain

and as ascending to 1,000 feet about the head of Anaktalak Bay; Blair (1943*a*: 11) adds Wabush Lake. According to Préfontaine and Cuerrier (1944: 106), it occurs in most rivers of the Gulf between Godbout River and Kecarpoui River. L. R. Richardson (1944: 207) records it from Lakes Mistassini and Wakonichi; and Allen (1948: 758, 762), from Blanc Sablon River. Legendre and Rousseau (1949: 134–135) present records from George River and Payne Lake, and ascribe *hudsonicus* to the brackish and salt waters of all Quebec. Munroe (1949: 166) places the brook trout in various waters of the Hamilton drainage (Wishart and Slimy lakes and Guy's River) and of the Koksoak drainage (Hanna's, Knob, Star, La Cosa, Snow, and Eclipse lakes and Star Creek); he mentions a weight up to 11½ pounds, indicates a spawning season after mid-October, and lists four parasites. Pomerleau (1950: 13) found it in the headwaters of the Manicouagan River. Dunbar and Hildebrand (1952: 95) include records from 24 miles north of Great Whale River on Hudson Bay, Payne Lake and River, Leaf River, Bobs Lake, and Koksoak River (up to 10 pounds); they mention "shrews and other small mammals" among the food. This species was found by Bateman (1953: 3) south of Leaf Bay, and by Oliver (1956; 1958) in Lac Aigneau and Astray Lake. Backus (1957: 292) presents records from many localities in the Atlantic drainage, from the Strait of Belle Isle north to Nain; they cover "sea-run" as well as non-migratory brook trout. McAllister and Bleakney (1959: 31) report the species from Seven Islands, Mile 134 (Q. N. S. & L. Railway), Slimy Lake, and Fort McKenzie and vicinity.

• *Leucichthys artedi* (Le Sueur). Lake herring; cisco or hareng de lac (Fr.); ?atshikoshmek (M.). Map 5.

Sebastien McKenzie (Plate 2, Fig. 3) told me at Lac Aulneau of a small kind of "whitefish"—different from the common whitefish and the round whitefish—that stays in small lakes and brooks and is known as "atshikoshmek" or "minkfish" because the mink ("tehékash") catches it. One of its known localities is a small lake near Fort McKenzie. P. Midlige (at Ashuanipi Lake) gave me an independent report of such a fish in a lake near Fort McKenzie. There would seem to be little doubt that this species is a *Leucichthys*.

Dunbar and Hildebrand (1952: 100) refer to Turner's report (MS., 1885) of two specimens taken from a lake trout's stomach near Fort Chimo, and they record a third specimen (181 mm.) of *artedi* from the same area. Dymond (1933: 8) lists specimens from about ten localities on the east side of James and Hudson bays, from Old Factory Bay in the south to Richmond Gulf in the north. L. R. Richardson (1944: 207) records specimens from Lake Wakonichi, including some found in the stomachs of lake trout. I know of no definite records from the Atlantic drainage or the St. Lawrence drainage in the Ungava Peninsula.

• *Coregonus atikameg atikameg* Bajkov. Hudson Bay whitefish. Map 6.

This small estuarine whitefish, which began to be noticed by explorers and naturalists as early as 1744, has had comparatively little attention from modern ichthyologists, and it received a technical name only a few years ago (Bajkov, 1933). Subsequently it was redescribed and figured by Fowler (1948: 148, Figs. 16–19), and I provided a historical sketch (1948: 161).

Bell (1879: 27) records, in rivers and brooks about the head of James Bay, "a species of whitefish (smaller than that of the great lakes)." He then adds (p. 28): "In the channels among the islands between East Main River and Cape Jones, the Indians were catching abundance of fish .... These consisted of the whitefish, above referred to, and a large and fine species resembling that of Lake Superior ...."

A further contribution to our knowledge of the present species comes from Low (1896: 329): "A small species of whitefish closely resembling the common whitefish is caught in abundance in the shallow salt water along the east coast of James Bay. These fish ascend the rivers of James Bay during the autumn months along with sea trout."

Low also states (1896: 86) that they ascend the East Main River from the sea, from September 1 to the freeze-up.

It is probably this same fish to which Joncas and Malouin refer (in Côté, 1933: 98): "At Smoky Hill Rapid [on Rupert River] during September, Indians of the region catch a species of small white fish which is very abundant there in that season. They catch thousands of these which are afterwards dried and smoked."

Anderson (1959: 80) also refers to these whitefish, as spawning at "Smoky Hills." This rapid is obviously not more than about 35 miles above the mouth of Rupert River, where falls of 102 feet are indicated on the Rupert House sheet (National Topographic Series).

From a little outside our territory, information is provided by Lower (1915), as reported by Dymond (1933: 5): "Lower in his account of the fish and fisheries of the west coast of

James Bay gives the average weight [of *Coregonus "clupeaformis"*=probably *C. a. atikameg* in this case] at not more than a pound and a quarter and sixteen inches long . . . . Larger specimens [perhaps *C. a. manitobensis* Bajkov] occur in inland lakes in the immediate vicinity."

Most of the specimens recorded by Dymond (1933: 3) as *C. clupeaformis* from Old Factory, Fort George, 25 miles up Big (Fort George) River, Little Cape Jones, and Great Whale River are perhaps the present estuarine form.

• *Coregonus clupeaformis* (Mitchill). Common whitefish; poisson blanc (Fr.); atikameg (M.). Map 7.

Since comparatively few gill-nets were set in the waters I visited, and since whitefish are not ordinarily taken by other means, only 25 specimens came to my attention. Three of these were taken in Sucker Creek; two in Lac Aulneau (Plate 1, Fig. 5); twelve in Mollie T. Lake; five in Ashuanipi Lake; and three in Ashuanipi River (Plate 1, Fig. 2). The species was also reported in Lac de Morhiban (Philippe Loth). At Eric Lake, Willé Pinette had been in the habit each fall of putting up several thousand fish (including whitefish) for winter use. Some of these localities are in the Ungava Bay drainage, some in the Hamilton River drainage, and others in the St. Lawrence drainage.

In the same part of Sucker Creek where a remarkable spawning aggregation of red suckers had been observed on June 11 and 12 (see account of that species), I found a school of about a dozen of the attending whitefish still lingering on June 24 and 25. Whether they were *Coregonus* or *Prosopium* is uncertain. Although the red suckers had evidently returned downstream to Abel Lake by that time, there were probably enough of their eggs remaining to attract the species preying upon them. During the evening of June 25 Ron Barrett reported a place about five feet square, perhaps a hundred yards higher up the creek, as covered with fishes of this sort. Out of a considerable number that he caught with a dipnet, he selected one large one (443 mm.) and two smaller ones (250 and 204 mm.) to bring to me. All three are *Coregonus clupeaformis*. The following color notes were made upon them after they had been in formalin overnight: dorsum dusky olive; sides silvery; venter and lower side of head white; fins dusky; pectorals and ventrals tinged with pale yellowish; dorsal, caudal, and anal with darker spots basally; lower margin of caudal and outer margin of ventrals whitish.

The two specimens from Lac Aulneau, 545 and 420 mm. in length (Plate 1, Fig. 5), were netted in water about 30 feet deep. Two females of about 385 and 375 mm., taken in Mollie T. Lake on August 13, contained eggs about 1 mm. in diameter. One taken in Ashuanipi River (Plate 1, Fig. 2) on August 29 was 527 mm. in length; it contained eggs about 1.5 mm. in diameter. Three specimens (440, 300, and 255 mm.), taken on August 30, Mile 216, Ashuanipi Lake, were netted near the surface 100 feet from shore in clear water 10 feet deep and with a rocky bottom.

On September 2 a fresh specimen from Ashuanipi River, about one mile below Ashuanipi Lake, was 348 mm. in length to tip of upper lobe of caudal; depth, 65 mm.; head, 59 mm. It furnished the following color notes: dorsum dusky, with silvery reflections; sides below lateral line silvery, with dusky punctulations except in the middle of the exposed portion of the scales; venter silvery white; iris silvery; preoperele and opercle silvery, with many dusky punctulations, and almost wholly dusky on upper portions; pectorals plain basally, somewhat dusky distally; other fins largely dusky.

E. H. Grainger reports insects in the stomach of the larger specimen from Lac Aulneau and insects, mollusks, and tapeworms in the stomachs of two adults from Mollie T. Lake. Acanthocephalan worms from the latter source have been identified as *Echinorhynchus* sp. by Aaron Goldberg, and the mollusks as *Valvata lewisi* Currier and *Pisidium casertanum* (Poli) by C. B. Wurtz.

Common whitefish occur quite generally in the lakes and streams of the forested parts of the Ungava Peninsula (see Map 7). Those recorded either as *C. clupeaformis* (Mitchill) or as *C. labradorius* (Richardson) (the latter name being generally considered a synonym of the former) are included in the following summation. The species is recorded by John Richardson (1836: 206) from "Musquaw [=Musquaro] River, that falls into the Gulf of St. Lawrence, near the Mingan Islands," this river being the type locality of *labradoricus;* by Low (1886: 7, 17) from Betsiamites River, Pipmuacan Lake, and Lake Mistassini and vicinity; further by Low (1896: 56, 63, 68, 70, 101, 137, 182, 329) from Lakes Obatogamau, Mistassini (up to 14 pounds), Nichicun, Winokapau, and Mushalagan; and again by Low (1898: 12, 15) from Clearwater and Natuakami lakes. Hamilton (1865: 113) mentions "Esquimaux Bay" [=Lake Melville]; Holme (1888: 195), Gull Island Lake, on Hamilton River; and Chambers (1896: 97, 115, 291), Lakes St. John, Manuan, and Wakonichi. Wallace (1907: 116) found the species in Kasheshebogamog Lake (northeast of Michi-

kamau Lake). Cabot (1912: 101, 154, 192) records it from the George River area, Assiwaban River, and all the lakes of the Davis Inlet area; Flaherty (1918: 120), from Lake Minto and the lower Leaf River; Merrick (1933: 39, 57), from Hamilton River above Minipi Rapid and from Big Lake near Grand Falls; and Blair (1943*a*: 11), from Muskrat Falls on Hamilton River and from Wabush Lake. Munroe (1949: 169) reports it from Knob Lake and as spawning about October 15. Dunbar and Hildebrand (1952: 101) state that it is common in most of the streams and lakes from Leaf River to George River. Additional localities are Flour Lake and McKenzie and Northwest rivers (Backus, 1957: 284), and Astray Lake and the outlet of Knob Lake (Oliver, 1958: 103).

• *Prosopium cylindraceum quadrilaterale* (Richardson). Round whitefish; menomini d'Amérique (Fr.); ?potmekosh (M.). Map 8.

On June 7 I had an opportunity to examine three fishes that had just been secured by Guy Giroux in Sucker Creek by means of a snatch hook. Unfortunately I had no preserving fluids at hand, but I judged the fishes to be *Prosopium,* and I have little doubt that they were. One had a total length of 387 mm.; depth, 71 mm.; head, 65 mm.; eye, 15.5 mm.; D. 12; A. 12; scales 10–87–8. (The number of scales in the lateral line suggests a probably significant difference from the local *Coregonus;* see Kendall, 1909: 209, Backus, 1957: 285, 286, and Fowler, 1961: 310. The dorsal coloration was like that of *Coregonus clupeaformis;* venter white. This and another individual of about equal size were females, each with small but clearly visible eggs. The third individual was a male, about 50 mm. shorter than the others. A workman present remarked that the Montagnais about Shabogamo Lake had a name for this species signifying "bonefish," because of its numerous bones.

On the same day, in the clear water just above the dam at the outlet of Knob Lake, I saw a round whitefish of 13 or 14 inches accompanying two smaller brook trout. It appeared yellowish olive on the dorsum and it had a series of faint parr marks along the sides, which proclaimed it a *Prosopium.*

On June 12, while observing a wonderful spectacle of hundreds of spawning red suckers in Sucker Creek, I noticed a considerable number of other fishes attending them in schools. While the suckers hugged the bottom, the others floated above their backs. I took them to be round whitefish, although they may have been, at least in part, common whitefish. Either species would doubtless have been attracted to the spot by the chance of feasting upon sucker eggs (see Bean, 1903: 228; Dunbar and Hildebrand, 1952: 102), although I detected none of them in the act. On June 27 Ron Barrett brought a 290-mm. *Prosopium* from the same part of Sucker Creek; it was probably taken in a dipnet.

Possibly the first publication of the vernacular name "shad-waiter" for *Prosopium* was by Prescott (1851: 342), when he bestowed upon the round whitefish of Lake Winnepesaukee, New Hampshire, the technical name of *Coregonus Nov-Angliae* (now relegated to the synonymy of *Prosopium cylindraceum quadrilaterale*). In those days shad (*Alosa*) ascended the Merrimac River and its tributaries as far as Lake Winnepesaukee to spawn. There they were presumably met and "waited upon" by round whitefish eager to devour their eggs. The subsequent construction of dams on the lower Merrimac excluded the shad from its upper waters. In like manner many of the other New England rivers have been rendered impassable for these fish (Bigelow and Schroeder, 1953: 110). So it might be difficult to say where the round whitefish of the present day are able to live up to their early reputation as "shad-waiters." There is little doubt, however, that in Ungava they play their role as "sucker-waiters."

Under the Montagnais name of "potmekosh," Sebastien McKenzie reported a fish at Fort McKenzie that I took to be the round whitefish.

At Mollie T. Lake on August 10 I was questioning Ben McKenzie on the food of the lake trout. In answer, he brought me the remains of a big individual caught six days previously, and took from its stomach a badly decomposed fish about 8 inches long. It was identifiable as *Prosopium* by a number of obscure dark spots (parr marks) on its sides. McKenzie said further that this species occurs by "thousands" in late September or early October in a creek connecting Hanna's and Peter's lakes, near the old Knob Lake airstrip. It may then be caught by hand in the shallow water, and it is utilized as bait for fur-bearing animals. This account was later confirmed by Jérôme St. Onge, who added that when the first snow falls, the fish retreat into the deep lakes. Norman Delmage spoke of once observing a "run" of these fishes in the above-mentioned creek when ice was forming in the fall. On September 30 I made a special trip

to this creek but found no fishes, either there or in a couple of near-by creeks.

A Montagnais boy, Michel Pinette, brought me a 178-mm. specimen from the Ashuanipi River a mile below Ashuanipi Lake. He seems to have caught it by hand on September 9, thus corroborating Ben McKenzie's previous account of this method of capture. After several years in preserving fluid, this specimen still exhibits about seven or eight faint parr marks above the lateral line; the posterior ones are more readily distinguishable than the anterior ones. These marks seem to be lost when the fish reaches a length of a foot or a little more.

There are comparatively few records of this species in the Ungava Peninsula. Préfontaine and Cuerrier (1944: 106) mention an individual from the Natashquan River and quote Hubbs as considering it an apparently undescribed subspecies. From the Atlantic drainage there are records by Kendall (1909: 209) from Northwest River; by Backus (1957: 286) from Mud, Little, and Flour lakes and Kenamu, Naskaupi, and English rivers; and by Oliver (1958: 103) from Astray Lake. Turner (MS., 1885: 728) describes a four-inch specimen from the Koksoak River; Dunbar and Hildebrand (1952: 103) report a 375-mm. specimen from the same river; Bateman (1953: 3), one from Irony Lake, south of Leaf Bay; and McAllister and Bleakney (1959: 31), two from Fort McKenzie. Hubbs and Lagler (1958: 56) give the species a range in eastern North America extending from Ungava Bay southward to Connecticut.

• *Mallotus villosus* (Müller). Capelin; capelan (Fr.); keskenemekosh (M.). Map 9.

The capelin plays a fairly important part in the economy of the coast of the Ungava Peninsula, its multitudes making up for its comparatively small size.

Spawning activity was evident at Seven Islands in late May and early June. It is said to last for three or four weeks. It seems to take place at high tide, in the shoal water on the sandy beach. Women and children were seen scooping up the fish with dipnets for family use. There is also a commercial fishery that employs seines that extend from the shore about 100 feet. The catch is made mostly at night. It is used for dog feed as well as for human consumption. (Much of this information was obtained from W. Foisy, of Labrador Fisheries, Ltd.)

On May 28 I noticed a dead individual in a meadow bordering the bay. On the next day several hundred were strung up to dry, with a line through their heads, by a house in town. They were pretty uniform in size, averaging perhaps about 8 inches in length. (According to Bigelow and Schroeder [1953: 134], "Few capelin are more than 6½ to 7½ inches long.")

On June 6, about midnight, Gilbert Simard observed vast numbers of capelins rolling in the edge of the bay at Seven Islands. He also spoke of many appearing at Thunder River, 70 miles to the east.

Cartwright (1792, 1: 148 and 2: 240 ff.) records capelins near Cape St. Charles and in Sandwich Bay. Cormack (1826: 32) reports on this species as food of, and bait for, cod on the shores of Labrador; according to his account, it spawns about the end of June and departs a month or more later. It was noted by Audubon on the southern coast from mid-July to early August (Storer, 1850: 265). Packard (1866: 275) gives July 12 as the date of the earliest appearance in great numbers at Square Island; he states that it occurs on the coast the year round but retreats to deep water in winter. It visits Occasional Harbour in August and September, being abundant by August 5 (Chimmo, 1868: 264). Bell (1879: 28) notes its occurrence on the east coast of Hudson Bay, apparently a little north of Cape Jones. Turner (MS., 1885: 724) found capelins in 1884 at the mouths of the George and Koksoak rivers. Townsend (1907: 22, 129) records the species off Bradore and at Battle Harbour. Wallace (1906: 45; 1907: 264–265) reports it as numerous in Hamilton Inlet about mid-July and as being shipped to northern Laborador for dog feed. Kendall (1909: 210; 1910: 508) records it from Chateau Bay, Northwest River, Greeley Harbour, and Egg Harbour. In early August Cabot (1912: 117) found the eggs lining the beaches, in some places 3 inches deep, north of Davis Inlet. In 1928 the schools were late in arriving on the North Shore of the Gulf and smaller than usual; hence the great black-backed gulls fed to a greater extent than usual on young American eiders (Lewis, 1928: 192). Another scarcity of capelins in 1932 resulted in considerable mortality among young gulls (Lewis, 1934: 98–99). Specimens from Saglek Bay, Nain, and Hawke Harbour are recorded by S. F. Hildebrand (1939: 5). Vladykov (1933: 8) had a specimen from Long Point, James Bay; he also (1946: 26, 60) reports the species as summer food of the white whale at the mouth of the Manicouagan River and at Les Escoumains. Templeman (1948: 9, map) mentions eleven localities, from Blanc Sablon in the south to Hebron in the north. Backus (1957: 295) presents records from Forteau Bay, Collingham's Cove, Emily Harbour, West Turnavik Island, and Hopedale Harbour.

The above accounts indicate that spawning takes place first on the North Shore of the Gulf and is progressively later at higher latitudes. As Tanner remarks (1947, 1: 432), the capelin "seems to be the decisive factor for the arrival of the cod. . . . Being the most selected food of the cod it is followed by the cod shoals."

• *Esox lucius* Linnaeus. Northern pike; brochet (Fr.); tshinosheo (M.). Map 10.

The Montagnais name is rendered by Lemoine (1901) as "tshinusheu."

Though widely distributed, the pike does not appear (from my own experience) to be a very common fish in the Ungava Peninsula. During the entire season just one specimen came to my notice. This was a female (Plate 1, Fig. 3), taken on a spinner at Guy's River on August 6 by Myron Kazmirchuk. Its total length was 740 mm.; its weight, 5 pounds; the ovaries were small and the stomach was empty. It provided these color notes on the following day: middorsum dark greenish; sides of head and body a slightly lighter green, with numerous elongate, pale greenish spots, becoming stripes on the caudal peduncle; also half a dozen such stripes extending from base of dorsal across upper part of body; lower parts, from chin to ventrals, whitish, changing to creamy from ventrals to caudal; fins dusky, variegated with pale greenish and tinged with reddish, the latter color restricted on the dorsal to its upper portion. Only the head was preserved.

The species was reported in Lac de Morhiban (Philippe Loth), and at Fort McKenzie on Swampy Bay River but not in Lac Aulneau (Sebastien McKenzie). A. E. Moss reported it in Howell's River, and Cyrille Dufresne in the Snelgrove Lake area. Willé Pinette puts up pike along with other species in the fall at his camp on Eric Lake. J. L. Véronneau (*in litt.*, June 26, 1955, and January 26, 1956) reports a number taken at the headwaters of the Nemiscau River in February, 1955, and at Lac Ochiltrie (up to 34 inches in length) in January, 1956.

Cartwright (1792, 2: 85, 199, 325) presents early records from Sandwich Bay and White Bear River. Hind (1863, 1: 198) records the pike from Lake Mushalagan, and Bell (1879: 27), from rivers and brooks about the head of James Bay. Low (1896: 17, 56, 63, 70, 86, 182, 331) considered it abundant throughout the interior in lakes and quiet streams, and common in rivers of the southern, eastern, and western watersheds, but less so in the Koksoak River; weight, 2 to 15 pounds. His localities include Lakes Obatogamau and Mistassini and East Main River. Chambers (1896: 12, 76, 104, 192, 193, 195, 206, 208, 235, 283, 284, 291) records the species in Lake St. John and its tributaries, in Lake Tschotagama (up to 49 pounds), in the Peribonca and Betsiamites rivers, Lac à Jim, and Pipmuacan and Big and Little Nekebau lakes. Wallace (1906: 123; 1907: 116) found it near Lake Disappointment and in Kasheshebogamog Lake. Additional localities are: Coxipi, Paradise, and upper St. Paul rivers (Cabot, 1920: 310); Flour Lake and Goose Bay (Merrick, 1933: 70); Wabush Lake (Blair, 1943*a*: 11); Lake Wakonichi (L. R. Richardson, 1944: 208); vicinity of Grand Falls (Desbarrats, 1948: 222); George River between 55° and 56° N. (Legendre and Rousseau, 1949: 134); Hanna's Lake and Lac La Cosa (Munroe, 1949: 169); Lac Bienville (Polunin, 1949: 47); English River at Baie Comeau (Wynne-Edwards, 1952: 19); Koksoak River, the George River post, and certain lakes in the Whale River drainage (Dunbar and Hildebrand, 1952: 107); McKenzie River and Muskrat Falls, Hamilton River (Backus, 1957: 296); Astray Lake (Oliver, 1958: 103); and Squaw Lake (McAllister and Bleakney (1959: 35). Legendre's map (1954: between pp. 40–41) indicates a distribution over the entire peninsula except for a narrow strip of the Atlantic seaboard.

• *Catostomus commersonii commersonii* (Lacepède). Common sucker, white sucker, gray sucker; carpe noire (Fr.); makatsheo (M.). Map 11.

Aside from the red sucker, the Montagnais distinguish another species of about the same size, under the name of "makatsheo." This is presumably *C. commersonii*. Ben McKenzie reported this species in Knob Lake and probably elsewhere in that area. "Gray suckers" are among the large numbers of fish that Willé Pinette puts up for winter use at Eric Lake.

This species is evidently much less common than the red sucker. It is reported from Lake Mistassini by Low (1896: 70, as "white sucker"); from the same lake and the neighboring Lake Wakonichi, with young 4 cm. long in August, by L. R. Richardson (1944: 207); from George River, between latitudes 55° and 57°, by Legendre and Rousseau (1949: 134); from Lake Mendry, near Fort Chimo, by Dunbar and Hildebrand (1952: 105); from the vicinity of Northwest River by Backus (1957: 297); and from Mile 134 (Q. N. S. & L. Railway) by McAllister and Bleakney (1959: 34).

• *Catostomus catostomus catostomus* (Forster). Red sucker; carpe, meunier de l'est (Fr.); mikoashi or mokoashi (M.). Map 12.

This species comes somewhat infrequently to the attention of fishermen who depend upon hooks rather than nets for their catch. An individual, with estimated length of 20 inches and weight of 3 pounds, was noticed on June 7 in still, shallow water just above the slight dam at the outlet of Knob Lake. On the following day four large ones were seen in Sucker Creek.

On June 10 a male (485 mm.) and a female (480 mm.) (Plate 2, Fig. 1) were snatch-hooked by Herb Chrysler in the outlet of Knob Lake. The next day they furnished the following color notes. Male: olive-green above, overlaid with dusky from insertion of dorsal to caudal; broad lateral stripe (maximum width above pectoral 1.5 inches), head to tail, bright rusty red; dorsal dusky; caudal and anal brownish, with numerous pearl organs; paired fins dull orange; lower sides pale brassy; venter white; head olive-green above, a little darker than back; lower side of head very pale yellowish orange; throat white; lips pale orange; eye with some golden. Female: dusky olive-green on back and sides, with very faint, obscure indication of a reddish lateral stripe; venter whitish; dorsal and caudal dusky, the latter dull orange on extreme lower portion; anal very pale orange; ventrals dull orange; pectorals dusky, dull orange distally; head dusky, the lower side whitish, washed with very pale orange; iris variegated with some golden. The gonads in both individuals were greatly enlarged; eggs were about 2.5 mm. in diameter. The stomach contents were very meager and not identifiable.

On June 11 Jean P. Labrecque reported about a hundred big suckers in Sucker Creek just above the right of way. On the following day there was a notable spawning aggregation at this spot, both above and below the culvert. They fairly covered the bottom of the creek, which is here perhaps two rods wide. The water was extremely clear, about one to two feet deep, with a rock and gravel bottom. One of three main schools above the culvert must have included about 400 fish. In general they faced upstream, and were so close that they almost touched each other's sides. Many lay almost inert, with a bare movement of the fins to maintain their position in the swift current; more evident exertion might have been expected of them. The brilliant red-sided males stood out among the more somber-colored females. Though I sat on the bank within five feet of the edge of the school, I was unable to make out any actual spawning. Now and then, at some disturbance, such as a passing truckman's tossing a rock into their midst, the fish were startled into disarray or disorganization, moving rapidly upstream and in part crosswise of the current; but they soon drifted back to their former position.

The masses of suckers were attended by a considerable number of somewhat smaller coregonids (either round whitefish or common whitefish, or both), floating over their backs in a higher stratum of the water and more or less forming schools of their own. Since this was not the spawning season for the latter, they were presumably attracted by the opportunity for preying upon the fresh spawn of the red suckers. One or more brook trout were also in attendance, perhaps with the same intent.

A motion-picture record of the spirited scene was made. It represented the greatest mass of spawning fish that I have ever observed. There must have been close to a thousand suckers. By June 25 they had disappeared from the creek.

On June 15 a school of perhaps fifty juvenile suckers appeared in the edge of Knob Lake over a rocky bottom where a small current entered through a culvert beneath a causeway. Four of them (the largest 38 mm.) were collected with a dipnet; presumably they had been spawned just a year previously. Another (35 mm.) was taken there in a minnow trap on June 21. Still another juvenile (38 mm.) was taken in the outlet cove on June 28.

On July 26 a 435-mm. individual (Plate 1, Fig. 5) was netted in Lac Aulneau in about 30 feet of water by Sebastien McKenzie, who said that strips of this species were good bait for lake trout. On August 24 Fred Farah scooped up a large number of juvenile individuals at the seaplane base at the north end of Ashuanipi Lake; they were about 10 feet from shore in clear water over a rocky bottom. Of 49 specimens preserved, the largest was 29 mm. in length. On August 28 Kom Pinette brought me an adult specimen (363 mm.) that he had found dead in Ashuanipi River below Ashuanipi Lake. It had marks on it which made him suspect that it was the victim of some predaceous bird, such as a herring gull (see Harper, 1953: 63). William Schrøpfer presented me with a 403-mm. specimen that he had taken in a gill-net in the same part of the river on September 2; the net had been set in five feet of water over a rocky bottom. Red suckers are among the fish that Willé Pinette puts up in the fall for use at his winter camp on Eric Lake.

Low (1896: 329) records suckers from this region under two different names: *Catostomus*

longirostris Le Sueur and *C. forsterianus* Richardson. Since both names are now regarded as synonyms of *C. catostomus*, it is difficult to interpret Low's records; it is quite possible that he had in hand *C. commersonii* as well as *C. catostomus*, although the former appears to be much the less common of the two species. On occasion he refers to these fishes merely as "suckers" or "carp." He states that his "red sucker" (*C. forsterianus*) is common throughout the interior, and that statement no doubt holds good today for the red sucker. Backus (1957: 296 fn.) is doubtless justified in considering that Low, when using the name *Catostomus longirostris*, was actually referring to the gray sucker (*C. commersonii*). Various other authors (Holme, 1888; Bryant, 1894; Wallace, 1907; Cabot, 1912; Munroe, 1949; and Polunin, 1949) do not particularize as to the species represented by their "suckers"; hence a citation of their records would be of dubious value. *C. catostomus* (or *C. c. catostomus*) is recorded from Northwest River by Kendall (1909: 209); from Lane's Bay by Cabot (1912: 213); from George River, between 55°01′ and 57°11′ N., by Legendre and Rousseau (1949: 134); from the Ungava Bay region—Leaf River and Fort Chimo—by Dunbar and Hildebrand (1952: 105); from south of Leaf Bay by Bateman (1953: 3); from McKenzie, Kenamu, Naskaupi, and English rivers, Mud, Little, Melville, and Flour lakes, and Kaipokok Fiord by Backus (1957: 296); from Astray Lake by Oliver (1958: 103); and from Mile 134 (Q. N. S. & L. Railway), Knob Lake airstrip, and Fort McKenzie by McAllister and Bleakney (1959: 33).

The dwarf subspecies, *Catostomus catostomus nanomyzon* Mather, has been reported from the North Shore of the Gulf—Godbout to Kecarpoui—by Préfontaine and Cuerrier (1944: 106) and from the Natashquan, Romaine, and Goynish rivers by Hubbs and Lagler (1958: 64).

• *Couesius plumbeus plumbeus* (Agassiz). Plumbeous minnow. Map. 13.

This species was first detected on June 7 just above the dam at the outlet of Knob Lake. Aside from a single fish seven or eight inches long, there were several schools, comprising half a dozen, a dozen, or 20 individuals of fairly uniform size (5 or 6 inches). Two of these schools combined in an assembly of 30 to 35 fish. For some time this larger group was accompanied by a four-inch brook trout as an integral part of it. In the limpid water (with a maximum depth of about two feet) some of the characters of the species were seen to fine advantage: a dusky lateral stripe, bordered above and below by orange (or yellowish?), and an orange spot at the base of the pectoral fin. When some of the fish approached the rapid at the outlet, they had to swim vigorously to keep from being swept downstream.

Three specimens (from 28 to 33 mm.) were taken on June 21 in a minnow trap in the edge of Knob Lake, over a rocky bottom. Two others (24 and 27 mm.) were collected on July 2 and 7 in a similar situation at the Iron Arm of Attikamagen Lake. On August 6 five specimens (from 83 to 110 mm.) were secured with a dip-net in Camp Brook. At the point of capture the brook is from 2 to 6 feet wide and from 2 to 8 inches deep, with a rocky to muddy bottom and a current varying from rapid to slow.

On August 24 two specimens (120 and 123 mm.) from the north end of Ashuanipi Lake were presented by Peter Midlige, who had taken them with a hook from a dock in five feet of fairly clear water, several rods from the shore. On the following day I saw two or three others at the same place. On September 4 I noted a dead individual along the Ashuanipi River a mile or so north of the lake. A final individual was seen on September 10 in Carol Lake.

Evidently this species has a wide distribution in the peninsula, although the records are comparatively few. Préfontaine and Cuerrier (1944: 106) report it from the rivers of the North Shore of the Gulf of St. Lawrence. L. R. Richardson (1944: 208) secured two specimens at Lake Wakonichi. Legendre and Rousseau (1949: 134) record it from 5 miles west of Indian House Lake. Backus (1951: 290) collected 21 specimens at Mud Lake (Hamilton River) and Northwest River; and later (1957: 297) he records further specimens from McKenzie River, Flour Lake, a tributary of Grand Lake, and Naskaupi River. The species has also been taken at Astray and Hanna's lakes, Foggy Lake (Geren's Mountain) (Oliver, 1958: 103), and at Knob Lake Airstrip and Fort McKenzie (McAllister and Bleakney, 1959: 34).

• *Anguilla rostrata* (Le Sueur). American eel; anguille d'Amérique (Fr.); oppimmishoi (M.). Map 14.

Sebastien McKenzie stated that he had caught an eel in the Moisie River about 50 years previously and that its Montagnais name is "oppimmishoi." This word is rendered by Lemoine (1901) as "upimishui."

Previous records of this species within the Ungava Peninsula are not numerous, and some of these indicate no exact locality. According to Hind (1863, 1: 298), it occurs along the North

Shore of the Gulf and furnishes the settlers with food. "The Indians report eels as common in the upper Romaine River" (Low, 1896: 332). Bigelow and Schroeder (1953: 154) include the Strait of Belle Isle and the northern side of the Gulf of St. Lawrence in the range of the species. Backus (1957: 298) records specimens from St. Mary's River and Mud Lake.

The occurrence of eels between Quebec and Montreal (Montpetit, 1897: 281–282, 294) is sufficient evidence of their passage through the Gulf of St. Lawrence. Further evidence comes from *The New York Times,* November 1, 1956: "For a distance of eighty miles downstream from Quebec City, on both shores of the St. Lawrence River, thousands of traps are holding back catches of eels . . . . Farmers—not fishermen—haul annually about 1,000,000 pounds of the black fish to a freezing plant." The antiquity of this industry is indicated by Kalm's observations (1937, 2: 423–435, 494) on eel traps along the St. Lawrence, both above and below Quebec City, in 1749.

It is interesting to find among the Montagnais at Lake St. John a taboo against eating eels, since they are considered a nonindigenous food (Speck, 1935: 80).

• *Gadus callarias* Linnaeus. Atlantic cod; morue commune (Fr.); ononoi (M.). Map 15.

On October 10 I noticed half a dozen codfish that had been split and laid out to dry on a rock near a house in Seven Islands. According to W. Foisy, of Labrador Fisheries, Ltd., these fish are caught locally in commercial quantities, but there was little activity in such operations in early October, the market being "down" (2 cents per pound fresh, 6 cents per pound dried). Several weeks previously the dominion government reportedly had been buying up codfish and shipping them to Greece for emergency relief. The local fishermen had a considerable quantity of dried cod left on their hands from the previous year. The drying fish are treated with very little salt.

Cartwright (1792, 1: *passim*) mentions an early fishery in the Cape St. Charles area. Low (1896: 332) reported the Atlantic cod as plentiful along the St. Lawrence and Atlantic coasts north to Cape Chidley, and along the eastern shore of Ungava Bay to the mouth of George River. It would appear, however, that the Ungava Bay records, at least for the most part, may have pertained actually to the Greenland cod (*Gadus ogac* Richardson) (see Dunbar and Hildebrand, 1952: 109). Vladykov (1933: 26) established the presence of *G. callarias* at Port Burwell; he also (1946: 26, 60) refers to it as the food of the white whale (*Delphinapterus leucas*) in summer near the mouth of the Manicouagan River and at Les Escoumains. Cabot (1912: *passim*) makes a number of references to the cod fishery in the Davis Inlet area. Grenfell (1922: 282–327) discusses historical, biological, and economic aspects of this fishery, with records from various localities along the coast from Cape Chidley to the Strait of Belle Isle. Thompson (1943) also presents a biological and economic study, with coastal records from the Gulf of St. Lawrence to Cape Chidley. Backus (1957: 298) has records along the east coast from Sandwich Bay to Nain. Other publications, for nearly a century past, have added comparatively little to the general picture of distribution along the southern and eastern coasts, as presented by Low.

• *Lota lota lacustris* (Walbaum). American burbot, ling; loche (Fr.); meli, meni, or miri (M.). Map 16.

Five immature specimens were taken in a minnow trap in Knob Lake by Guy Giroux, June 22 to July 6; they varied in length from 78 mm. on the first date to 118 mm. on the last date. One of them appears in Plate 2, Figure 2. The trap was set in shallow water in the edge of the lake, on a rocky bottom. Three individuals of approximately the same size had been captured there previously and used for trout bait before I had a chance to examine them. The foregoing specimens had probably been hatched a little more than a year previously (see McCrimmon, 1959: 449). A 16-mm. larva was collected on July 12 at the mouth of a creek on the Iron Arm, Attikamagen Lake. Sebastien McKenzie reported none of this species in Lac Aulneau.

Low (1896: 332) reports the ling, with a weight of 2 to 15 pounds, as common in all the deep lakes throughout the interior and as an important source of food for the Indians. There are few records from specific localities: Lake Mistassini (Bignell, 1885: 18; Low, 1890: 19); Lake St. John (Chambers, 1896: 11, 288); Assiwaban River (Cabot, 1912: 154); Minipi Lake (Cabot, 1920: 310); Fort Chimo (Dunbar and Hildebrand, 1952: 109); McKenzie River, Mud Lake, and a tributary of Flour Lake (Backus, 1957: 299); Astray Lake (Oliver, 1958: 103); and Mile 134 (Q. N. S. & L. Railway) (McAllister and Bleakney, 1959: 35).

• *Gasterosteus aculeatus* Linnaeus. Three-spined stickleback; épinoche à trois épines (Fr.); ?tsistaskoannamesh (M.). Map 17.

This diminutive fish appears to be quite common and generally distributed in the interior lakes. It is readily observed in the shoal waters that it frequents. I found it in Attikamagen, Aulneau, and Mollie T. lakes, and saw single specimens, doubtless of this species, in Knob and Al's lakes. A few were seen in the Iron Arm of Attikamagen Lake, and many more in the Northwest Bay; in the latter area, on July 17, there were schools of a dozen, fifty, or even a hundred individuals. Seven specimens (from 31 to 65 mm.) were taken with a dipnet in Attikamagen Lake from July 2 to 17; they were in clear, shallow water near shore, over a rocky bottom. Some had a bright orange-red stripe on the back of the ventral spines. In others (adult as well as immature) this stripe was faint or wanting; it may be correlated with sex or season rather than with age. Among three specimens taken in Lac Aulneau on July 27 and August 2, the stripe was present in a small one (36 mm.) but lacking in the largest (53 mm.). Bean remarks (1903: 341) that the dorsal spines "may be fixed and immovable at the will of the fish." Occasionally I noticed this condition in the ventral spines, as if they were locked in position at right angles to the body. It might be considered a defense reaction against predaceous animals.

On July 24 (and probably for a day or two previously) sticklebacks fairly swarmed in the shoal waters at a sandy and gravelly point on Lac Aulneau. They were in schools of as many as 100 individuals or perhaps more; nearly all, with the exception of half a dozen or so, appeared smaller than those in Attikamagen Lake. On July 25, however, I could not detect a single individual at this locality. Possibly their disappearance was correlated with a change of weather. On the 22nd and 23rd it had been sunny, and on the latter day the temperature ranged from 68° to 85°F. On the 24th hard rain came, with a drop in temperature from 63° to 49.5°. Thereafter, until August 2, the weather was constantly cloudy or rainy, with an average daily minimum temperature of about 43° and a maximum of about 49.5°. Late on August 2 the sun came out and the temperature rose to 54°; at the same time a dozen or more sticklebacks appeared, whereas on only one of the preceding eight days—July 27—had I noted just a few. Perhaps they had sought deeper water offshore during the cool, rainy spell. Bigelow and Welsh (1925: 170) speak of the species "dropping down into slightly deeper water such as the bottoms of the deeper creeks to pass the cold months."

If the Montagnais name supplied at the beginning of this account actually pertains to the present species, it would seem appropriate enough, for its meaning, according to Sebastien McKenzie, is "fish with a nail." The "nail" is presumably the ventral spine.

Most of the previous records (under the specific or subspecific names of *aculeatus* or *cuvieri*) from the Ungava Peninsula are from salt or brackish coastal waters. These range from Wakeham Bay on Hudson Strait (Vladykov, 1933: 10) and Cape Hope's Advance, Chimo, and George River (Dunbar and Hildebrand, 1952: 111) to the east coast: Red Bay (Storer, 1850: 254); Chateau Bay, Hamilton Inlet, Rigolet, Lake Melville, Northwest River, Battle and Indian harbours (Kendall, 1909: 211, and 1910: 508); and Forteau Bay (Backus, 1957: 300). Records from interior waters are few: Surnock Pond (S. F. Hildebrand, 1939: 7); Payne Lake (Legendre and Rousseau, 1949: 134); Bobs Lake near the George River trading post (Dunbar and Hildebrand, 1952: 111); south of Leaf Bay (Bateman, 1953: 3); McKenzie and Naskaupi rivers and Flour Lake (Backus, 1957: 300); Astray Lake (Oliver, 1958: 103); and Fort McKenzie (McAllister and Bleakney, 1959: 36). Curiously enough, there seem to be no records for the North Shore of the Gulf, except at Salmon River in extreme eastern Saguenay County; yet the range extends south to New Jersey.

• *Stizostedium vitreum* (Mitchill). Wall-eyed pike, pike-perch; doré jaune (Fr.). Map 18.

J. L. Véronneau (*in litt.*, June 26, 1955) wrote me of a "doré" taken during the previous January out of a lake at the headwaters of the Nemiscau River. Men in his party (fishermen from the Gaspé) apparently knew it by that name. When I sent him an outline sketch of this species (from Legendre, 1954: fig. 78), he confirmed the identification.

The range of this species in the Ungava Peninsula seems to be restricted to the James Bay drainage and to the western part of the St. Lawrence drainage. The following localities are on record for it: Lake Mistassini, Rupert and East Main rivers, tributaries of Lake St. John, and Betsiamites River (rare in that river and not found to the eastward) (Low, 1886: 17; 1896: 70, 332); Peribonca River, Obatogamau Lake, Lac à Jim, Big and Little

Nekebau lakes, and Lake Wakonichi (Chambers, 1896: 192, 208, 209, 235, 291); and 25 miles up Big [=Fort George] River (Vladykov, 1933: 13). Cabot (1920: 305; 1922: 206) extends the range considerably to the east of the limit assigned by Low—not only "as far as the eastern heads of the Maniquagan," but even "farther, well toward Minipi," a lake tributary to the lower Hamilton River. Legendre (1954: map between pages 68 and 69) indicates a distribution north only to the Ashuapmuchuan River and east only to the Saguenay River.

• *Cottus cognatus* Richardson. Great Bear Lake sculpin; chabot (Fr.). Map 19.

Sculpins are apparently well distributed in the interior. Eight specimens were obtained in Attikamagen, Knob, and Carol lakes. On June 28 two individuals were detected by Guy Giroux some 6 to 10 feet offshore in Knob Lake, as they skulked among rocks in a foot or two of water. One had a generally dark and somewhat uniform coloration, while the other exhibited dark bars on a lighter ground color. They eluded capture; but a specimen was taken there in a minnow trap by Guy Giroux on July 4. Another was seen on September 23 just above the dam at the outlet of this lake.

James R. Murdoch captured a specimen by hand on July 2 close to the shore at Northwest Bay, Attikamagen Lake. Another was taken there in the same manner on July 18. Four others were collected (mostly by Antoine Deschênes) with a dipnet in the Iron Arm of this lake on July 7 and 12; they were near the shore in shallow water with a rocky bottom.

The following color notes were made on an adult of July 7: head with numberless minute punctulations and fewer only slightly larger black dots, giving the head as a whole a dusky appearance; anterior dorsum, to end of first dorsal, similar to head; from this point to caudal about a dozen dusky blotches of irregular size and shape; membranes of all fins with extremely fine dusky punctulations; dorsal and anal rays tinged with extremely pale orange; upper border of dorsal also pale orange, but a little more pronounced; this tinge barely suggested on rays of pectorals, ventrals, and caudal.

Two subadults, captured on July 12 and kept alive in an aquarium jar, provided the following notes: ground color very pale buffy, marked with dusky spots of various sizes and shapes; most of the fins with the faintest tinge of very pale orange on rays (almost lacking on anal); upper border of first dorsal a little brighter; iris with a narrow golden rim.

The sculpins of Attikamagen Lake displayed much the same habits as those of the Canadian Northwest. They hug the bottom and move a short distance at a time, coming to rest again on outspread pectorals; they remain thus for considerable periods and are almost never seen to progress, while undisturbed, more than a couple of inches above the bottom. Perhaps they are not quite so prone as their western congeners to hide beside or beneath rocks.

At Carol Lake a sculpin was detected near shore among the rocks of the bottom on September 9, and another was collected there on September 17.

The taxonomy of the genus *Cottus* is a study of peculiar difficulty; it has been interpreted variously and perhaps none too satisfactorily. The forms of the Ungava Peninsula have been recorded under a number of different names: as *C. cognatus* Richardson and *C. gracilis* Heckel from Northwest River (Kendall, 1909: 212, 213); as *C. cognatus* from south of Leaf Bay (Bateman, 1953: 3) and from Squaw Lake, Fort McKenzie, and Fort Chimo (McAllister and Bleakney, 1959: 36); as *C. bairdii bairdii* Girard from Lake Wakonichi (L. R. Richardson, 1944: 208), from Lake St. John (Hubbs and Lagler, 1947: 97), and from a tributary of Mud Lake and from Little Lake, Northwest River (Backus, 1951: 292); as *C. bairdi kumlieni* (Hoy) from Flour Lake, Naskaupi River, and tributaries of Grand Lake (Backus, 1957: 310); and as *C. cognatus gracilis* "Häckel" from the Koksoak River and its tributaries (Dunbar and Hildebrand, 1952: 114) and from McKenzie River and vicinity, Little Lake, and Flour Lake (Backus, 1957: 311). The distribution of the various forms of *Cottus* in the peninsula, as indicated in the literature, is shown on Map 19.

• *Cyclopterus lumpus* Linnaeus. Lumpfish. Map 20.

On May 27 a dead individual, about nineteen inches in length, was found on the shore of Seven Islands Bay. A sketch and descriptive notes leave no doubt as to its identity. According to Bigelow and Schroeder (1953: 460), the maximum length recorded from the American coast is 23 inches.

Storer (1850: 269) reports the species at Bras d'Or and Red Bay; Turner (MS., 1885: 708), at Rigolet; Vladykov (1946: 69), at Les Escoumains (as food of the white whale); Bigelow and Schroeder (1953: 459), in the Gulf of St. Lawrence and on the outer coast of Labrador; and Backus (1957: 322), in Hamilton Inlet and at West Turnavik Island.

LITERATURE CITED

ALLEN, ARTHUR A. 1948. Sea bird cities off Audubon's Labrador. Nat. Geog. Mag., June: 755–774, 7 pls., 3 figs., 1 map.

ANDERSON, WILLIAM ASHLEY. 1959. The angel of Hudson Bay. The true story of Maude Watt and her courageous husband, who saved a people from extinction. Saturday Evening Post **231** (January 24, 1959): 24, 80–82, 3 figs.

BACKUS, RICHARD H. 1951. New and rare records of fishes from Labrador. Copeia **1951** (4): 288–294.

———. 1957. The fishes of Labrador. Bull. Am. Mus. Nat. Hist. **113** (art. 4): 273–337, 2 pls., 2 figs.

BATEMAN, BOB. 1953. Observations on the natural history of the Leaf Bay–Fort Chimo region, Ungava, Quebec. Intermediate Naturalist **8**: 1–7, 1 pl.

BEAN, TARLETON H. 1903. Catalogue of the Fishes of New York. New York State Mus. Bull. 60. 1–784.

BELL, ROBERT. 1879. Report on an exploration of the east coast of Hudson's Bay, in 1877. Geol. Survey Canada Rept. Progress for 1877–8: 1C–37C, 6 pls., 3 figs.

———. 1886. Observations on the geology, zoology, and botany of Hudson's Strait and Bay, made in 1885. Geol. and Nat. Hist. Survey Canada Ann. Rept. (n. s.) 1: 1DD–27DD, 2 pls., 3 figs., 1 map, "1885."

BIGELOW, HENRY B., AND WILLIAM C. SCHROEDER. 1953. Fishes of the Gulf of Maine. Rev. ed. U. S. Fish and Wildlife Service, Fishery Bull. 74. viii + 577, 288 figs.

BIGELOW, HENRY B., AND WILLIAM W. WELSH. 1925. Fishes of the Gulf of Maine. Bull. U. S. Bur. Fisheries 40, pt. 1. 1–567, 278 figs.

BIGNELL, F. H. 1885. Notes of a journey to Lake Mistassini. Trans. Geog. Soc. Quebec **1** (4): 13–25.

BLAIR, A. A. 1943*a*. Salmon Investigations, 1. Obstructions in Newfoundland and Labrador Rivers. Newfoundland Govt. Dept. Natural Resources Research Bull. 12 (fisheries): 3–50, 8 maps.

———. 1943*b*. Salmon Investigations, 2. Atlantic Salmon of the East Coast of Newfoundland and Labrador, 1939. Newfoundland Govt. Dept. Natural Resources Research Bull. 13 (fisheries): 1–21, 6 figs., 1 map.

BLEAKNEY, SHERMAN. 1954. Food habits of lake trout from Squaw Lake, northern Quebec. Canadian Field-Naturalist **68** (2): 94.

BOUTHILLIER, C. DE B. 1948. Report of the Committee on Fish. Appendix 1. Prov. Quebec Assoc. Protection Fish and Game Ann. Rept.: 24–26.

BRYANT, HENRY G. 1894. A journey to the Grand Falls of Labrador. Geog. Club Philadelphia Bull. **1** (2): 37–86, 8 pls., 2 maps.

———. 1913. Explorations in southeastern Labrador. Bull. Geog. Soc. Philadephia **11** (1): 1–15, 10 fig., 1 map.

CABOT, WILLIAM B. 1912. In Northern Labrador. Boston. xii + 292, 48 pls., 2 maps.

———. 1920. Labrador. London. xiii + 354, 48 pls., 2 maps.

———. 1922. The Indians. In: Wilfred T. Grenfell and others, Labrador, the Country and the People. Rev. ed. 184–225, 2 pls.

CARTWRIGHT, GEORGE. 1792. A Journal of Transactions and Events, during a Residence of Sixteen Years on the Coast of Labrador . . . . Newark [England]. **1**: xvi + [6] + 287, 1 pl., 2 maps; **2**: x + 505; **3**: x + 248 + 15.

CAYLEY, EDWARD. 1863. Up the River Moisie. Trans. Liter. and Hist. Soc. Quebec, n. s., **1**, pt. 1: 75–92.

CHAMBERS, E. T. D. 1896. The Ouananiche and its Canadian Environment. New York. xxii + 357, 15 pls.

[CHIMMO, WILLIAM.] 1868. A visit to the northeast coast of Labrador, during the autumn of 1867, by H.M.S. "Gannet," Commander W. Chimmo, R. N. Jour. Royal Geog. Soc. **38**: 258–281, 1 map.

COMEAU, NAPOLEON A. 1923. Life and Sport on the Lower St. Lawrence and Gulf. 2d ed. Quebec. 1–440, 16 pl., 3 figs., 1 map. (Orig. ed., 1909.)

CORMACK, W. E. 1826. On the natural history and economic uses of the cod, capelin, cuttle-fish, and seal, as they occur on the banks of Newfoundland, and the coasts of that island and Labrador. Edinburgh New Philos. Jour. **1**: 32–41.

CÔTÉ, GEORGES. 1933. Annual report of the director of surveys. Rept. Minister Lands and Forests Prov. Quebec 1932, appendix 22: 91–130.

COUPER, WM. 1868. Investigations [in 1867] of a Naturalist between Mingan and Watchicouti, Labrador. Quebec. 1–14.

DESBARRATS, G. H. 1948. Surveying on the Hamilton River, Labrador. Canadian Geog. Jour. **37** (5): 214–229, 18 figs., 2 maps.

DUNBAR, M. J., AND H. H. HILDEBRAND. 1952. Contribution to the study of the fishes of Ungava Bay. Jour. Fisheries Research Board Canada **9** (2): 83–128, 1 fig.

DURGIN, GEORGE FRANCIS. 1908. Letters from Labrador. Concord, N. H. 1–117, 8 pls.

DYMOND, J. R. 1933. Biological and oceanographic conditions in Hudson Bay. 8. The coregonine fishes of Hudson and James Bays. Contrib. Canadian Biol. and Fisheries, n. s., **8** (1): 1–12.

———. 1941. Atlantic salmon in Ungava Bay. Canadian Field-Naturalist **55** (2): 19–20.

———. 1947. A List of the Freshwater Fishes of Canada East of the Rocky Mountains, with Keys. Royal Ontario Mus. Zool. Misc. Publ. 1. 1–56.

EVERMANN, BARTON WARREN, AND EDMUND LEE GOLDSBOROUGH. 1907. A check list of the freshwater fishes of Canada. Proc. Biol. Soc. Washington **20**: 89–119.

EVERMANN, BARTON WARREN, AND WILLIAM CONVERSE KENDALL. 1902. An annotated list of the fishes known to occur in the St. Lawrence River. Rept. U. S. Fish Comm. **27**: 227–240.

FLAHERTY, ROBERT J. 1918. Two traverses across

Ungava Peninsula, Labrador. Geog. Rev. **6** (2): 116–132, 12 figs., 1 map.

FORTIN, PIERRE. 1862. [Cetacea, fishes, etc., of Canadian shores of Gulf of St. Lawrence.] Ann. Rept. 1862, appendix: 109–124.

———. 1863. Continuation. Ibid., 1863: 60–72.

———. 1864. Continuation. Ibid., 1864: 61–69.

———. 1865. Continuation. Ibid., 1865: 67–79.

FOWLER, HENRY W. 1948. Fishes of the Nueltin Lake Expedition, Keewatin, 1947. Part 1—Taxonomy. Proc. Acad. Nat. Sci. Philadelphia **100**: 141–152, 25 figs.

———. 1961. Taxonomic notes on fishes of the interior of the Ungava Peninsula. Jour. Elisha Mitchell Sci. Soc. **77** (2): 309–311, 2 figs.

GATHORNE-HARDY, G. M. 1911. On fishing in the Labrador. In: H. Hesketh Prichard, Through Trackless Labrador (New York): 221–231.

GRENFELL, WILFRED T., AND OTHERS. 1922. Labrador, the Country and the People. New ed. New York. xxvi + 529, 47 pls., 19 figs., 6 maps. (Orig. ed., 1909).

HAMILTON, R. V. 1865. On the portion of the coast of Labrador between Blanc Sablon Bay, in lat. 51°20′ N., and Cape Harrison, in lat. 55° N. Proc. Royal Geog. Soc. **9**: 131–137.

HARPER, FRANCIS. 1948. Fishes of the Nueltin Lake Expedition, Keewatin, 1947. Part 2—Historical and field notes. Proc. Acad. Nat. Sci. Philadelphia **100**: 153–184, 2 pls.

———. 1953. Birds of the Nueltin Lake Expedition, Keewatin, 1947. Am. Midland Naturalist **49** (1): 1–116, 8 figs., map.

———. 1958. Birds of the Ungava Peninsula. Univ. Kansas Mus. Nat. Hist. Misc. Publ. 17. 3–171, 6 pls., 26 maps.

———. 1961. Land and Fresh-Water Mammals of the Ungava Peninsula. Univ. Kansas Mus. Nat. Hist. Misc. Publ. 27. 1–178, 8 pls. 3 figs. 45 maps.

HILDEBRAND, HENRY. 1949. Notes on an abundance of shrews (*Sorex cinereus cinereus*) and other small mammals in the Ungava Bay region of far northern Quebec. Jour. Mammalogy **30** (3): 309–311.

HILDEBRAND, SAMUEL F. 1939. An annotated list of the fishes collected on the several expeditions to Greenland, the Fox[e] Basin region, and the coast of Labrador by Capt. R. A. Bartlett, from 1925 to 1935. Meddelelser om Grønland **125** (1): 1–12.

HIND, HENRY YOULE. 1863. Explorations in the interior of the Labrador Peninsula, the country of the Montagnais and Labrador Indians. London. **1**: xvi + 351, 9 pls., 15 figs., 1 map; **2**: xv + 304, 4 pls., 7 figs., 1 map.

HOLME, RANDLE F. 1888. A journey in the interior of Labrador, July to October, 1887. Proc. Royal Geog. Soc., n. s., **10** (4): 189–205, 1 map.

HUBBARD, MINA BENSON. 1908. A Woman's Way through Unknown Labrador. London. xvi + 338, 50 pls., 1 map.

HUBBS, CARL L., AND KARL F. LAGLER. 1947. Fishes of the Great Lakes Region. Cranbrook Inst. Sci. Bull. 26. 1–186, 26 pls., 250 figs., 38 diagrams, 2 maps.

———. 1958. Fishes of the Great Lakes Region. Rev. ed. Cranbrook Inst. Sci. Bull. 26 xiii + 213, 77 pls., 49 figs., 1 chart, 2 maps.

JORDAN, DAVID STARR. 1929. Manual of the Vertebrate Animals of the Northeastern United States, Inclusive of Marine Species. 13th ed. Yonkers-on-Hudson. xxxi + 446, 15 figs., 1 map.

JORDAN, DAVID STARR, BARTON WARREN EVERMANN, AND HOWARD WALTON CLARK. 1930. Check List of the Fishes and Fishlike Vertebrates of North and Middle America North of the Northern Boundary of Venezuela and Colombia. Rept. U. S. Commission Fisheries 1928, appendix 10: 1–670.

KALM, PETER. 1937. Peter Kalm's Travels in North America. (The English version of 1770 revised from the original Swedish and edited by Adolph B. Benson.) Vol. II. New York. 381–797, 3 pls., 1 map.

KENDALL, WILLIAM CONVERSE. 1909. The fishes of Labrador. Proc. Portland Soc. Nat. Hist. **2** (8): 207–243.

———. 1910. Report on the fishes collected by Mr. Owen Bryant on a trip to Labrador in the summer of 1908. Proc. U. S. Nat. Mus. **38** (1763): 503–510, 1 pl.

LEGENDRE, VIANNEY. 1954. Key to the Game and Commercial Fishes of the Province of Quebec. The Freshwater Fishes. Vol. I. First English ed. Prov. Quebec Game and Fisheries Dept., Montreal. 1–180, 80 figs., numerous diagrams, 3 maps.

LEGENDRE, V., AND JACQUES ROUSSEAU. 1949. La distribution de quelques-uns de nos poissons dans le Québec arctique. Annales Assoc. Canadienne-Française Av. Sci. **15**: 133–135.

LEMOINE, GEO. 1901. Dictionnaire français-montagnais . . . . Boston. 1–281, 1–63.

LEWIS, HARRISON F. 1928. Notes on birds of the Labrador Peninsula in 1928. Canadian Field-Naturalist **42** (8): 191–194.

———. 1934. Notes on birds of the Labrador Peninsula in 1931, 1932 and 1933. Canadian Field-Naturalist **48** (6): 98–102; (7): 115–119.

LOW, A. P. 1886. Report of the Mistassini Expedition, 1884–5. Geol. and Nat. Hist. Survey Canada Ann. Rept. (n. s.) 1: 1D–55D, 1 map, "1885."

———. 1890. The Mistassini region. Ottawa Naturalist **4**: 11–28.

———. 1896. Report on explorations in the Labrador Peninsula along the East Main, Koksoak, Hamilton, Manicuagan and portions of other rivers, in 1892–93–94–95. Geol. Survey Canada Ann. Rept., n. s., 8, 1895: 1L–387L, 2 pls.

———. 1898. Report on a traverse of the northern part of the Labrador Peninsula from Richmond Gulf to Ungava Bay. Geol. Survey Canada Ann. Rept. (n. s.) 9: 1L–43L, 4 pls.

LOWER, A. R. M. 1915. A report on the fish and fisheries of the west coast of Hudson Bay. Ann.

Rept. Naval Service Canada 1914 (appendix): 29–67.

LUCAS, FREDERIC A. 1891. Explorations in Newfoundland and Labrador in 1887, made in connection with the cruise of the U. S. Fish Commission schooner Grampus. Rept. U. S. Nat. Mus. 1888–89: 709–728, 1 map.

MCALLISTER, D. E., AND SHERMAN BLEAKNEY. 1959. Some freshwater fishes from northeastern Quebec and Labrador. Nat. Mus. Canada Bull. **166**: 31–39, 1 map.

MCCARTHY, EUGENE. 1894. The Leaping Ouananiche, What It Is, Where, When and How to Catch It. New York. 1–66, illus.

MCCRIMMON, HUGH R. 1959. Observations on spawning of burbot in Lake Simcoe, Ontario. Jour. Wildlife Management **23** (4): 447–449.

MCLEAN, JOHN. 1932. John McLean's Notes of a Twenty-Five Year's Service in the Hudson's Bay Territory. Ed. W. S. Wallace. Publ. Champlain Soc. 19. Toronto. xxxvi + 402, 1 map. (Orig. ed., 1849.)

MARTIN, N. V. 1955. Limnological and biological observations in the region of the Ungava or Chubb Crater. Jour. Fisheries Research Board Canada **12** (4): 487–496, 2 figs., 2 maps.

MERRIAM, C. HART, VERNON BAILEY, E. W. NELSON, AND E. A. PREBLE. 1910. Fourth provisional zone map of North America. In: Check-list of North American Birds, prepared by a committee of the American Ornithologists' Union. 3d. ed. Frontispiece. New York.

MERRICK, ELLIOTT. 1933. True North. New York. [5] + 353, 8 pls., 1 map.

MONTPETIT, A. N. 1897. Les poissons d'eau douce du Canada. Montreal. xiv + 553, 14 pls., 215 figs., 1 map.

MUNROE, EUGENE G. 1949. Notes on fish of the interior of the Labrador Peninsula. Arctic **2** (3): 165–173, 3 figs., 1 map.

OBALSKI, J. 1905. Explorations in the Lake Chibogamau region. In: Mining Operations in the Province of Quebec for the Year 1904. 5–21, 5 pls. Dept. Lands, Mines, and Fisheries, Quebec.

OLIVER, D. R. 1956. Limnological investigations in northern Quebec. Arctic **8** (4): 261, "1955."

———. 1958. A limnological investigation in the Knob Lake area. McGill Sub-Arctic Research Laboratory Ann. Rept. 1956–57, suppl. 1: 101–105.

PACKARD, ALPHEUS SPRING, JR. 1866. List of vertebrates observed at Okak, Labrador, by Rev. Samuel Weiz, with annotations by A. S. Packard, Jr. Proc. Boston Soc. Nat. Hist. **10**: 264–277.

———. 1891. The Labrador Coast. New York. 1–506, 5 pls., 35 figs., 13 maps.

POLUNIN, NICHOLAS. 1949. Arctic Unfolding. London. 1–348, 33 pls., 3 maps.

POMERLEAU, RENÉ. 1950. Au sommet de l'Ungava. Revue Université Laval **4** (9): 1–16, 2 maps.

POWER, G. 1958. Salmon investigations on the Koksoak River, 1957. Arctic **11** (1): 63–64.

PRÉFONTAINE, G., AND J.-P. CUERRIER. 1944. Notes sur quelques poissons du Comté de Saguenay. Annales Assoc. Canadienne-Française Av. Sci. **10**: 106.

PRESCOTT, WILLIAM. 1851. Descriptions of new species of fishes; from a "Synopsis of the fishes of the Winnipiseogee and its connecting waters." Am. Jour. Sci. and Arts (ser. 2) **11**: 340–345.

PRICHARD, H. HESKETH. 1910. Across Labrador from Nain to the George or Barren Ground River. Geog. Jour. **36** (6): 691–692, 2 pls.

RICHARDSON, JOHN. 1836. Fauna Boreali-Americana . . . . Part 3. The Fish. London. xv + 327, 24 pls.

RICHARDSON, L. R. 1944. Brief record of fishes from central northern [more properly, southwestern] Quebec. Copeia **1944** (4): 205–208.

SCOTT, W. B. 1954. Freshwater Fishes of Eastern Canada. Toronto: xiv + 128, 110 figs., 1 map.

SLASTENENKO, E. P. 1958. The Freshwater Fishes of Canada. Toronto. 1–388, 138 figs., 1 map.

SPECK, FRANK G. 1935. Naskapi: the Savage Hunters of the Labrador Peninsula. Norman, Okla. 3–248, 20 pls., 28 figs., 1 map.

STEARNS, W. A. 1883. Notes on the natural history of Labrador. Proc. U. S. Nat. Mus. **1883**: 111–137.

STORER, HORATIO ROBINSON. 1850. Observations on the fishes of Nova Scotia and Labrador, with descriptions of new species. Boston Jour. Nat. Hist. **6**: 247–270, 2 pls.

TANNER, VÄINÖ. 1947. Outlines of the Geography, Life & Customs of Newfoundland-Labrador. Vol. I. Cambridge. 1–436, 342 figs.

TEMPLEMAN, WILFRED. 1948. The Life History of the Caplin in Newfoundland Waters. Bull. Newfoundland Govt. Laboratory 17 (research). 1–151, 1 pl., 29 figs., 7 maps.

THOMPSON, HAROLD. 1943. A Biological and Economic Study of Cod (*Gadus callarias*, L.) in the Newfoundland Area. Newfoundland Govt. Natural Resources Research Bull. 14 (fisheries). 1–160, 3 figs., 23 maps.

TOWNSEND, CHARLES WENDELL. 1907. Along the Labrador Coast. Boston. i–xii, 11–289, 26 pls., 1 map.

———. 1913. A short trip into the Labrador Peninsula by way of the Natashquan River. Bull. Geog. Soc. Philadelphia **11** (3): 170–182, 4 pls.

TURNER, LUCIEN M. 1885. Fishes of the Ungava District, Labrador. Unpublished manuscript. U. S. National Museum Library.

VLADYKOV, VADIM D. 1933. Biological and oceanographic conditions in Hudson Bay. 9. Fishes from the Hudson Bay region (except the Coregonidae). Contrib. Canadian Biol. and Fisheries **8** (2): 13–49, 1 fig., 4 maps.

———. 1946. Études sur les mammifères aquatiques. IV.—Nourriture du Marsouin Blanc ou Béluga (*Delphinapterus leucas*) du fleuve Saint-Laurent. Contrib. Dept. Pêcheries Québec 17: 5–155, 1 pl., 39 figs., 3 maps.

———. 1949. Quebec lampreys (Petromyzontidae).

1.—List of Species and their Economical Importance. Prov. Quebec Dept. Fisheries Contrib. 26: 2–67, 1 pl., 21 figs.

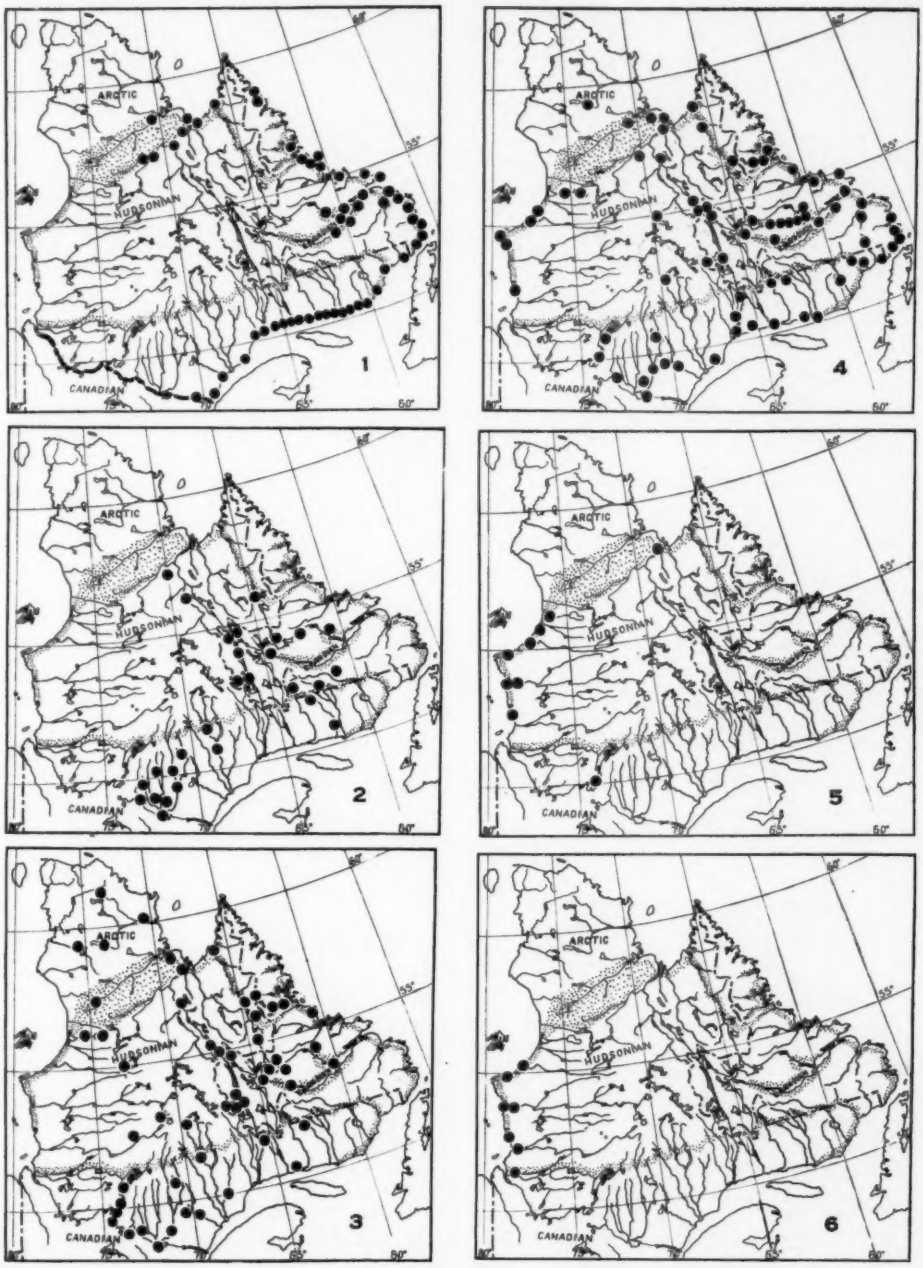
WALLACE, DILLON. 1906. The Lure of the Labrador Wild. 6th ed. New York. 1–339, 16 pls., 3 maps.

———. 1907. The Long Labrador Trail. New York. xii + 315, 27 pls., 2 maps.

WEED, ALFRED C. 1934. Notes on the sea trouts of Labrador. Copeia **1934** (3): 127–133.

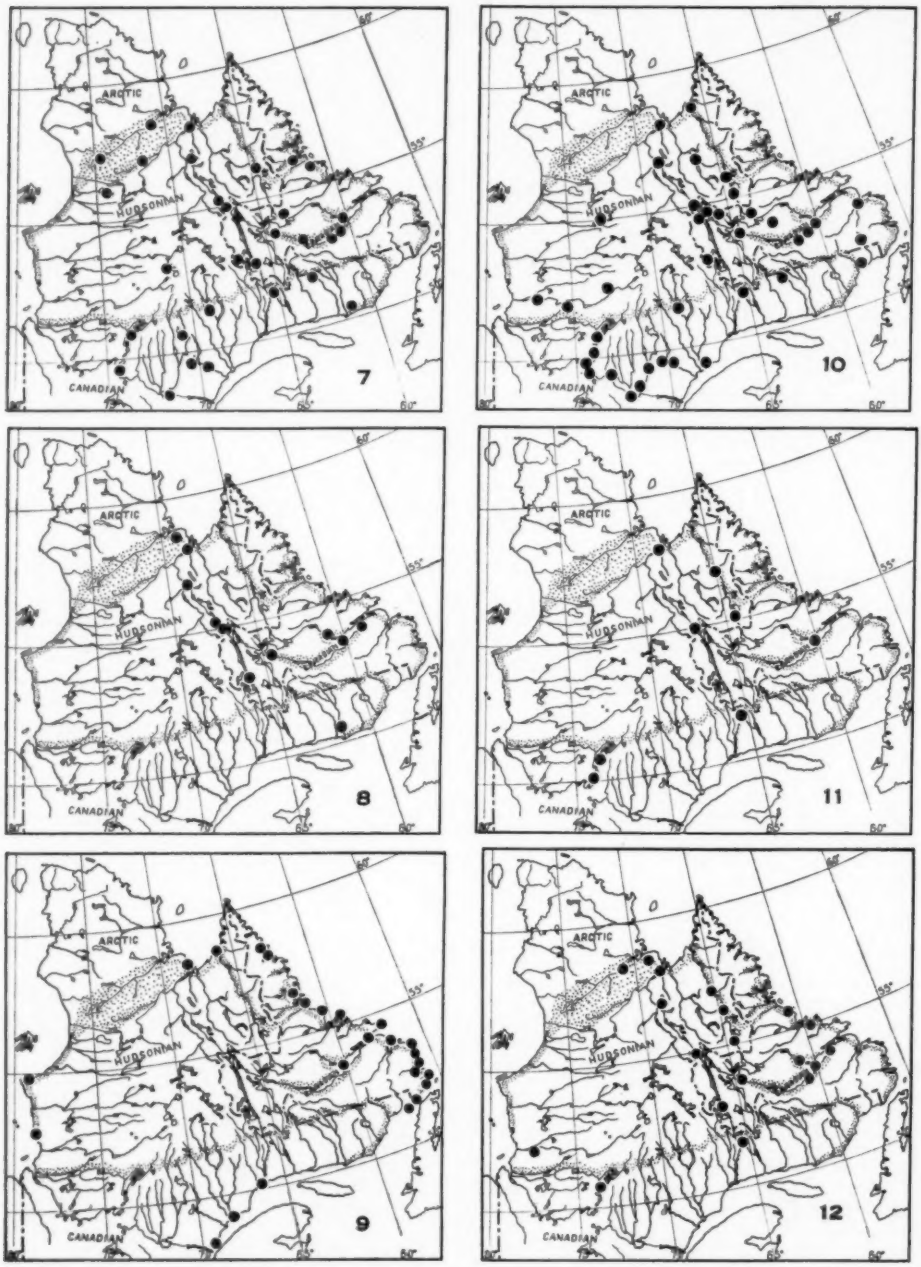
WEIZ, SAMUEL. 1866. List of vertebrates observed at Okak, Labrador, by Rev. Samuel Weiz, with annotations by A. S. Packard, Jr. Proc. Boston Soc. Nat. Hist. **10**: 264–277.

WYNNE-EDWARDS, V. C. 1952. Freshwater vertebrates of the Arctic and Sub-Arctic. Fisheries Research Board Bull. 94: [iii] + 28, 2 figs., 1 map.



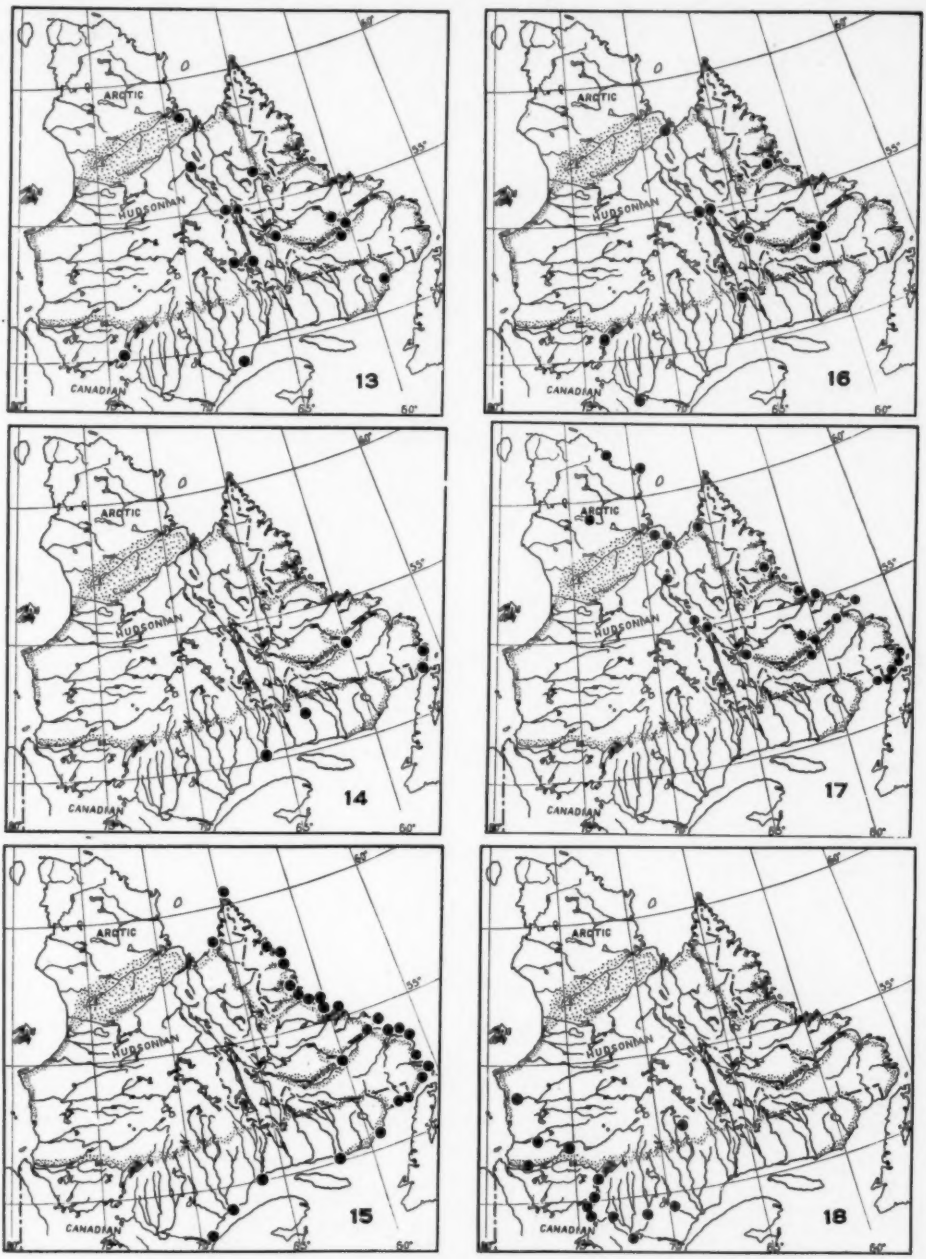
Distributional Records of Fishes in the Ungava Peninsula

Map 1. *Salmo salar salar.*
Map 2. *Salmo salar ouananiche.*
Map 3. *Cristivomer namaycush.*
Map 4. *Salvelinus fontinalis.*
Map 5. *Leucichthys artedi.*
Map 6. *Coregonus atikameg atikameg.*



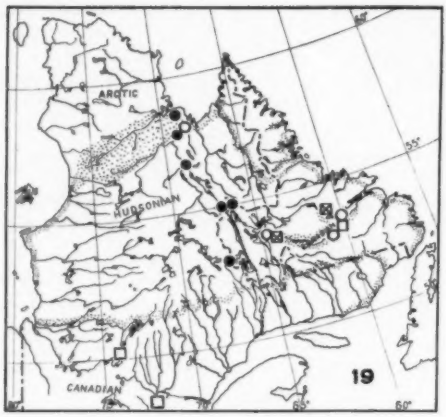
Distributional Records of Fishes in the Ungava Peninsula

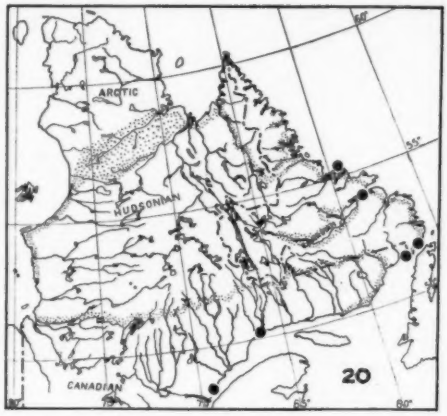
Map 7. *Coregonus clupeaformis.*
Map 8 *Prosopium cylindraceum quadrilaterale.*
Map 9. *Mallotus villosus.*
Map 10. *Esox lucius.*
Map 11. *Catostomus commersonii commersonii.*
Map 12. *Catostomus catostomus catostomus.*



Distributional Records of Fishes in the Ungava Peninsula

Map 13. *Couesius plumbeus plumbeus.*
Map 14. *Anguilla rostrata.*
Map 15. *Gadus callarias.*
Map 16. *Lota lota lacustris.*
Map 17. *Gasterosteus aculeatus.*
Map 18. *Stizostedium vitreum.*





Distributional Records of Fishes in the Ungava Peninsula

Map 19.

- ● *Cottus cognatus (cognatus).*
- ○ *Cottus cognatus gracilis.*
- □ *Cottus bairdii bairdii.*
- ⊠ *Cottus bairdii kumlieni.*

Map 20. *Cyclopterus lumpus.*

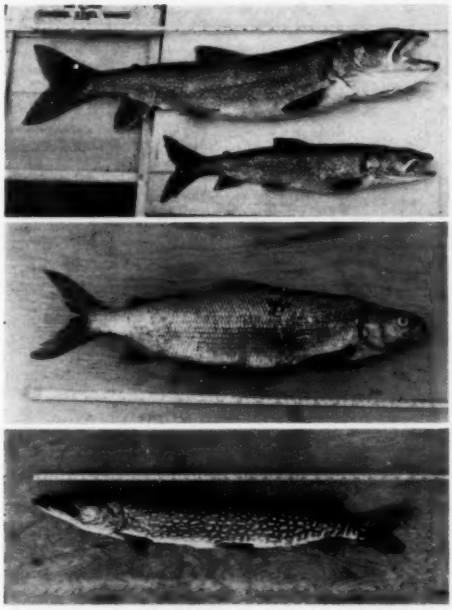


FIG. 1. Lake trout (*Cristivomer namaycush*), the larger about 747 mm. Carol Lake, Labrador, September 12, 1953. FIG. 2. Common whitefish (*Coregonus clupeaformis*), female, 527 mm. Ashuanipi River, 2 miles north of Ashuanipi Lake, Labrador, August 29, 1953. FIG. 3. Northern pike (*Esox lucius*), female, 740 mm. Guy's River, Labrador, August 6, 1953.

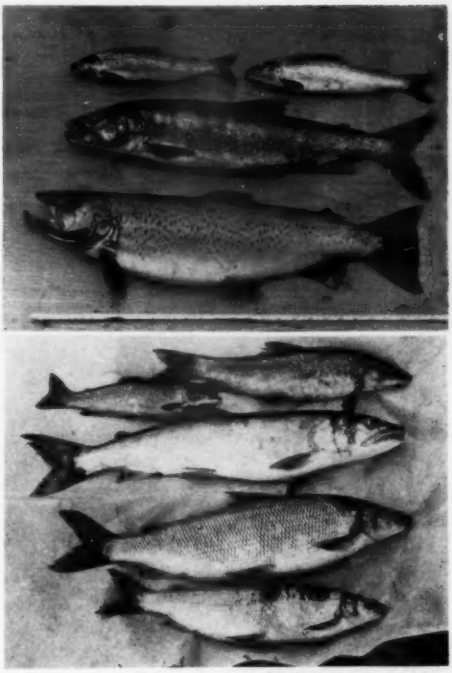


FIG. 4. Lake trout (*Cristivomer namaycush*) (1, center: 615 mm.) and ouananiche (*Salmo salar ouananiche*) (3: 278, 315, and 640 mm.). Ashuanipi River, 2 miles north of Ashuanipi Lake, Labrador, August 29, 1953. FIG. 5. Red sucker (*Catostomus catostomus catostomus*) (1: 435 mm.); brook trout (*Salvelinus fontinalis*) (1: 290 mm.); lake trout (*Critivomer namaycush*) (1: female, 615 mm.), and common whitefish (*Coregonus clupeaformis*) (2: 545 and 420 mm.). Lac Aulneau, Quebec, July 26, 1953.

PLATE 1.

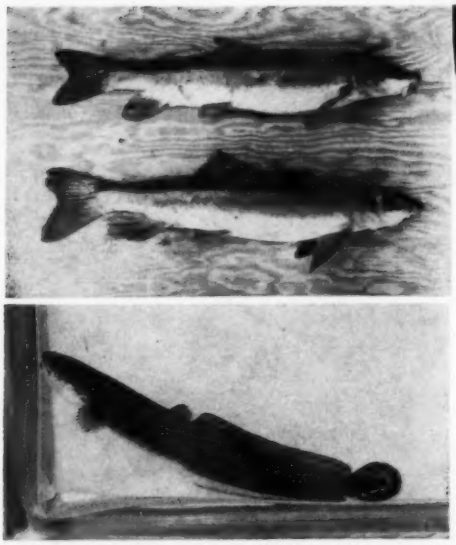


FIG. 1. Red suckers (*Catostomus catostomus catostomus*): female (above), 480 mm.; male (below), 485 mm. Outlet of Knob Lake, Quebec, June 10, 1953. (Note pearl organs on caudal and anal fins of male.) FIG. 2. American burbot (*Lota lota lacustris*), 81 mm. Knob Lake, Quebec, June 23, 1953.

FIG. 3. Sebastien McKenzie (Montagnais) cleaning a lake trout. Lac Aulneau, Quebec, July 26, 1953. FIG. 4. Fishing for ouananiche in Ashuanipi River, 2 miles north of Ashuanipi Lake, Labrador (William Bazuk and James Thompson). September 6, 1953.

PLATE 2.

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Chapel Hill, N. C.

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(5) BEAUER, A., AND N. MACON. Amer. Jour. Math. **71**: 349–361. 1949.

BAILEY, JOSEPH R., AND DAVID G. FREY. 1951. Darters of the genus *Hololepis* from some natural lakes of North Carolina. Jour. Elisha Mitchell Sci. Soc. **67**: 191–203.

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